Research Square

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Low cyclosporine concentrations in children and time to acute graft versus host disease

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Abstract

Background

Achievement of target blood concentrations of cyclosporine (CsA) early after transplantation is known to be highly effective for reducing the incidence of acute graft versus host disease (aGVHD). However, no research has been conducted for predicting elapsed time from low CsA concentration to aGVHD occurrence. The objective of this study was to investigate the risk of aGVHD according to elapsed time from low CsA concentrations in children with allogenic hematopoietic stem cell transplantation (HSCT).

Methods

The outcomes of 61 consecutive children who underwent allogeneic HSCT and received CsA as prophylaxis against aGVHD between May 2012 and March 2015 were retrospectively evaluated; the main outcome was any association between elapsed time from low CsA concentrations to aGVHD occurrence. Elapsed time was defined as follows: one week (0–6 days), two weeks (7–13 days), or three weeks (14–20 days), and the incidence of aGVHD was examined for the first three months after transplantation.

Results

In children whose mean CsA concentration after the allogenic HSCT did not reach the initial target concentration, the adjusted odds ratio for developing aGVHD within one week was 12.3 (95% confidence interval [CI]: 2.7–56.0). The AORs for developing aGVHD between 7 and 13 days and between 14–20 days were 90.8 (95% CI: 8.3–999.9) and 28.1 (95% CI: 2.4–333.1), respectively.

Conclusions

Careful attention needs to be paid to the elapsed time after low CsA concentrations are detected to prevent aGVHD.

Introduction

Allogeneic hematopoietic stem cell transplantation (HSCT) is an important treatment method for many hematologic malignancies, bone marrow dysfunctions, immunodeficiency diseases, and metabolic diseases [1, 2]. However, the long-term survival after allogenic HSCT is hindered by the development of human leukocyte antigen and the occurrence of graft-versus-host disease (GVHD). Thus, preventing and treating GVHD are important for reducing morbidity and mortality [3, 4].

Corticosteroids, cyclophosphamide, and antithymocyte globulin have long been used to prevent GVHD, and cyclosporine-A (CsA) as an immunosuppressive agent was introduced in the late 1970s. Since then, CsA has been used with methotrexate (MTX) or methylprednisolone. Recently, the combination of CsA and MTX has been used as a standard preventive therapy for acute GVHD (aGVHD) [5, 6].

CsA is an 11 amino acid residue belonging to the group of cyclopeptides isolated from Tolypocladium inflatum Gams; it inhibits the early cellular immune response to stimulation and has a T-cell-specific inhibitory effect as well. CsA also binds to a cyclophilin receptor protein to form a heterodimeric complex and inhibits the dephosphorylation of nuclear factor of activated T cells by binding to calcineurin, which acts as a transcription factor for the interleukin-2 gene [7]; in other words, CsA binding to cyclophilin inhibits calcineurin activity and suppresses calcineurin-induced cascade. In addition, CsA increases the expression of transforming growth factor-β, thereby inhibiting the production of cytotoxic T cells and contributing to immunosuppressive activity [8, 9].

Although CsA has been widely used, it is difficult to predict its blood concentrations because of its high pharmacokinetic variability. Moreover, the narrow therapeutic range of CsA requires close monitoring after drug administration [10, 11]. Findings from a number of studies have suggested that low CsA concentrations increase the risk of aGVHD and have shown a correlation between trough CsA concentrations and aGVHD incidence. In particular, researchers have reported that reaching target blood concentrations of CsA early after transplantation is highly effective for lowering the incidence of aGVHD [12–14]; however, there has been no research on predicting elapsed time from low CsA concentration to aGVHD occurrence. Therefore, the purpose of this study was to investigate the risk of aGVHD according to elapsed time, in weeks, from low cyclosporine concentrations in children who underwent allogenic HSCT.

Methods

Study patients

We conducted this retrospective observational study with patients who underwent allogenic HSCT and received CsA from the pediatrics department of Asan Medical Center in Seoul, Korea, from April 2012 to March 2015; we excluded patients with a previous history of transplantation and those who were older than age 18. The study was approved by the Asan Medical Center Institutional Review Board (IRB number: 2017 – 0509).

The collected data were age, sex, body weight, diagnosis, dates of transplantation and engraftment, use of voriconazole, levels of serum creatinine (SCr), aspartate aminotransferase, and alanine aminotransferase, and donor type (sibling, matched unrelated, or mismatched unrelated). We also analyzed the use of busulfan, cyclophosphamide, fludarabine, antithymocyte globulin, and total body irradiation as conditioning regimens and use of MTX and mycophenolate mofetil as concomitant therapy with CsA for preventing aGVHD. In addition, we classified renal function according to the National Cancer Institute Criteria for Adverse Events (NCI CTCAE) based on SCr the day before transplantation.

CsA administration

CsA was administered intravenously at a rate of 3 mg/kg/day with a 12-hr interval from the day before transplantation and was converted to oral dosing after the blood concentration reached a stable target

range; patients received an oral dose of CsA (soft capsule) twice daily. We measured trough CsA blood concentrations at least three times per week; the target concentrations were 105–155 ng/mL for patients with sibling donors and 155–210 ng/mL for those with other donor types.

We measured CsA blood concentrations from day 0 to day 30 based on the transplantation day, and the main outcome was any association between elapsed weeks from low CsA concentrations to aGVHD occurrence. We defined elapsed time in weeks as follows: one week (0–6 days), two weeks (7–13 days), or three weeks (14–20 days). We also checked for incidence of aGVHD in the first three months after transplantation.

Statistical analysis

We used the chi-squared test or Fisher's exact test to compare the categorical variables between patients with and without aGVHD and used multivariable logistic regression analysis to identify independent risk factors for aGVHD; we included in the multivariate analysis factors that showed $p < 0.05$ in the univariate analysis along with strong confounding with age and sex. We calculated odds ratios and adjusted odds ratios (AOR) from univariate and multivariate analyses, respectively, and considered $P < 0.05$ statistically significant. We performed all statistical analyses using SPSS version 17.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results

Among 63 eligible patients for this study, we excluded two, one for a previous history of transplantation and one for age > 18. Accordingly, we used the data from 61 pediatric patients for the analysis; Table 1 presents the baseline characteristics of those 61 patients.

Table 1

Clinical characteristics of patients (n = 61)

Characteristics	No (%) or Mean ± SD	aGVHD No (%) or Mean ± SD		p
		Absence (n = 34)	Presence (n = 27)	
Age (years)				0.166
< 12	37 (60.7%)	18 (52.9%)	19 (70.4%)	
≥ 12	24 (39.3%)	16 (47.1%)	8 (29.6%)	
Sex				0.980
Female	27 (44.3%)	15 (44.1%)	12 (44.4%)	
Male	34 (55.7%)	19 (55.9%)	15 (55.6%)	
Body weight (kg)	33.8 ± 20.0	37.2 ± 9.5	29.5 ± 20.1	0.138
Diagnosis				0.228
ALL	16 (23.2%)	7 (20.0%)	9 (33.3%)	
AML	22 (36.1%)	14 (40.0%)	8 (29.6%)	
SAA	11 (18.0%)	4 (11.4%)	7 (25.9%)	
MDS	7 (11.5%)	7 (20.0%)	1 (3.7%)	
Others	5 (8.2%)	3 (8.6%)	2 (7.4%)	
Donor type				0.331
Sibling	22 (36.1%)	15 (44.1%)	7 (25.9%)	
Mismatched unrelated	13 (21.3%)	6 (17.6%)	7 (25.9%)	
Full matched unrelated	26 (42.6%)	13 (38.2%)	13 (48.1%)	
Busulfan				0.980
Yes	34 (55.7%)	19 (55.9%)	15 (55.6%)	

rATG: rabbit anti-thymocyteglobulin, TBI: total body irradiation, MTX: methotrexate, AST: aspartate aminotransferase, ALT: alanine transferase, NCI CTCAE: National Cancer Institute Common Terminology Criteria for Adverse Events, CsA: cyclosporine

One week: 0–6 days elapsed after low cyclosporine concentration detected; two weeks: 7–13 days elapsed after low cyclosporine concentration detected; three weeks: 14–20 days elapsed after low cyclosporine concentration detected

Characteristics	No (%) or Mean ± SD	aGVHD No (%) or Mean ± SD		p
No	27 (44.3%)	15 (44.1%)	12 (44.4%)	
Cyclophosphamide				-
Yes	61 (100.0%)	34 (100.0%)	27 (100.0%)	
No	0 (0.0%)	0 (0.0%)	0 (0.0%)	
Fludarabine				0.027
Yes	31 (50.8%)	13 (38.2%)	18 (66.7%)	
No	30 (49.2%)	21 (61.8%)	9 (33.3%)	
rATG				0.048
Yes	32 (52.5%)	14 (41.2%)	18 (66.7%)	
No	29 (47.5%)	20 (58.8%)	9 (33.3%)	
TBI				0.222
Yes	16 (26.2%)	11 (32.4%)	5 (18.5%)	
No	45 (73.8%)	23 (67.6%)	22 (81.5%)	
MTX				0.041
Yes	48 (78.7%)	30 (88.2%)	18 (66.7%)	
No	13 (21.3%)	4 (11.8%)	9 (33.3%)	
Voriconazole				0.685
Yes	6 (9.8%)	4 (11.8%)	2 (7.4%)	
No	55 (90.2%)	30 (88.2%)	25 (92.6%)	
AST/ALT				0.735
< 200	51 (83.6%)	29 (85.3%)	22 (81.5%)	
≥ 200	10 (16.4%)	5 (14.7%)	5 (18.5%)	
NCI CTCAE				0.008

rATG: rabbit anti-thymocyteglobulin, TBI: total body irradiation, MTX: methotrexate, AST: aspartate aminotransferase, ALT: alanine transferase, NCI CTCAE: National Cancer Institute Common Terminology Criteria for Adverse Events, CsA: cyclosporine

One week: 0–6 days elapsed after low cyclosporine concentration detected; two weeks: 7–13 days elapsed after low cyclosporine concentration detected; three weeks: 14–20 days elapsed after low cyclosporine concentration detected

Characteristics	No (%) or Mean ± SD	aGVHD No (%) or Mean ± SD		p
0–1	50 (82.0%)	32 (94.1%)	18 (66.7%)	
2–5	11 (18.0%)	2 (5.9%)	9 (33.3%)	
Week reached initial target CsA concentration				0.098
0	2 (3.3%)	2 (5.9%)	0 (0.0%)	
1	16 (26.2%)	10 (29.4%)	6 (22.2%)	
2	33 (54.1%)	20 (58.8%)	13 (48.1%)	
3	7 (11.5%)	1 (2.9%)	6 (22.2%)	
4	3 (4.9%)	1 (2.9%)	2 (7.4%)	
Initial target concentration reached before engraftment				0.155
Yes	44 (72.1%)	27 (79.4%)	17 (63.0%)	
No	17 (27.9%)	7 (20.6%)	10 (37.0%)	
Weeks elapsed after low CsA concentration				
1 week				< 0.001
Yes	28 (45.9%)	8 (23.5%)	20 (74.1%)	
No	33 (54.1%)	26 (76.5%)	7 (25.9%)	
2 weeks				< 0.001
Yes	20 (37.0%)	3 (8.8%)	17 (85.0%)	
No	34 (63.0%)	31 (91.2%)	3 (15.0%)	
3 weeks				0.007
Yes	17 (38.6%)	9 (26.5%)	8 (80.0%)	
No	27 (61.4%)	25 (73.5%)	2 (20.0%)	

rATG: rabbit anti-thymocyteglobulin, TBI: total body irradiation, MTX: methotrexate, AST: aspartate aminotransferase, ALT: alanine transferase, NCI CTCAE: National Cancer Institute Common Terminology Criteria for Adverse Events, CsA: cyclosporine

One week: 0–6 days elapsed after low cyclosporine concentration detected; two weeks: 7–13 days elapsed after low cyclosporine concentration detected; three weeks: 14–20 days elapsed after low cyclosporine concentration detected

The median age of the study population was 10.0 years (range: 0.7–18.0), and the median body weight was 31.3 kg (range: 7.4–77.7); 44.3% of patients were female. Acute myelogenous leukemia was the most common disease (22 patients, 36.1%), followed by acute lymphocytic leukemia (16 patients, 23.2%), severe aplastic anemia (11 patients, 18.0%), and myelodysplastic syndrome (7 patients, 11.5%). The donor type proportions were 36.1% siblings, 21.3% mismatched unrelated, and 42.6% matched unrelated, and the median number of engraftment days was 11 (range: 9–22).

The incidence of aGVHD was 0.27 times lower in patients with MTX (P = 0.041) and 3.2 times higher in patients with fludarabin therapy (P = 0.027). Patients with NCI CTCAE grades 2 or higher had an 8-fold greater incidence of aGVHD (P = 0.008); specifically, the rates were 13.8, 58.6, and 11.1 times higher at elapsed times of one week, two weeks, and three weeks, respectively, after the mean CsA concentrations did not reach therapeutic concentrations (Tables 1 and 2).

Table 2

Univariate and multivariate logistic regression analysis to identify predictors of acute GVHD related to cyclosporine administration

Characteristics	Unadjusted OR (95% CI)	Adjusted OR (95% CI)		
		Model I	Model II	Model III
Age < 12	0.474 (0.163–1.375)	1.412 (0.295–6.767)	4.002 (0.366–43.743)	5.197 (0.465–58.128)
female	0.987 (0.357–2.729)	1.479 (0.371–5.903)	1.260 (0.196–8.090)	0.997 (0.177–5.601)
Fludarabine	3.231 (1.122–9.303)*	1.976 (0.440–8.866)	4.497 (0.547–36.959)	3.369 (0.627–18.100)
MTX	0.267 (0.072–0.993)*	0.680 (0.098–4.703)	0.885 (0.064–12.288)	0.436 (0.045–4.239)
NCI CTCAE 0–1	8.000 (1.556–41.134)*	9.694 (1.403–66.990)*	1.847 (0.187–18.265)	
Weeks elapsed after low CsA concentration detected				
1 week	9.286 (2.882–29.917)***	12.284 (2.698–55.933)*		
2 weeks	58.556 (10.632–322.499)***		90.832(8.251–999.903)***	
3 weeks	11.111 (1.976–62.466)**			28.086 (2.368–333.090)

Model I included age, sex, MTX use, fludarabine therapy, NCI CTCAE grade 2 or higher, and elapsed time of one week. Models II and III included elapsed times of two and three weeks, respectively.

MTX: methotrexate, NCI CTCAE: National Cancer Institute Common Terminology Criteria for Adverse Events, CsA: cyclosporine, OR: odds ratio

One week: 0–6 days elapsed after low cyclosporine concentration detected; two weeks: 7–13 days elapsed after low cyclosporine concentration detected; three weeks: 14–20 days elapsed after low cyclosporine concentration detected

*P < 0.05, **P < 0.01, ***P < 0.001

We constructed multivariate analysis models to determine independent factors for aGVHD occurrence in respective elapsed times of one, two, and three weeks. Model I included age, sex, MTX use, fludarabine therapy, NCI CTCAE grades 2 or higher and elapsed time of one week, and Models II and III included elapsed times of two and three weeks, respectively. When the mean CsA concentration after allogenic HSCT did not reach the initial targets, the AOR for developing aGVHD within one week was 12.3 (95% CI: 2.7–56.0). The AORs for developing aGVHD between 7 and 13 days and between 14 and 20 days were 90.8 (95% CI: 8.3–999.9, Model II) and 28.1 (95% CI: 2.4–333.1, Model III), respectively. In Model I, patients with NCI CTCAE grades 2 or higher had 9.7 times greater incidence of aGVHD.

Discussion

aGVHD is an important complication of allogenic HSCT; authors have reported frequencies of up to 80%. aGVHD occurs by stimulating the immune system of the host, resulting in damage to organs such as the skin, liver, and gastrointestinal tract.

CsA, which is used to prevent aGVHD after a patient receives allogenic HSCT, leads to higher incidence and greater severity of aGVHD in low rather than high doses, but some authors have reported that low doses reduce the recurrence of blood cancer [15, 16]. Therefore, determining the appropriate dose of CsA is important.

CsA blood concentrations can change with lower metabolic rates or depending on CsA excretion rates, which depend on conditions such as renal or liver function. Particularly in pediatric patients, high doses are required to maintain blood concentrations [17] because of their higher distribution volumes and elimination rates compared with those of adults. In addition, appropriate therapeutic concentrations of CsA are affected by conditioning regimens and concomitant medications, and thus there is still much controversy regarding the appropriate treatment concentrations and timing for preventing aGVHD.

Recent study authors have reported that high CsA concentrations within three to four weeks after transplantation are more effective in preventing aGVHD. For instance, Garcia et al. [18] reported a correlation between CsA concentration and aGVHD in an adult patient with 156 allogenic HSCTs; low CsA at three weeks after transplantation increased the risk of severe aGVHD. Kanda et al. [19] examined the effect of CsA on 171 adult allogenic HSCT patients and found that the CsA concentration within three weeks after transplantation was the most important factor in determining the risk of severe aGVHD. On the contrary, other researchers have reported that CsA concentrations in the first two weeks after transplantation were significantly related to aGVHD in pediatric transplant recipients, and similarly, in some studies, low CsA concentrations during the first two weeks after transplantation in adult transplant recipients increased the risk of aGVHD [12–14].

In contrast to the fact that many study findings suggest that CsA blood concentrations should reach a therapeutic range in the initial stages after transplantation, there is no study of the risk of developing aGVHD based on the time elapsed after the CsA concentration did not reach the therapeutic range; thus, we quantified this risk based on this elapsed time. Given that one third or more of the study patients did

not reach the therapeutic range at one or two weeks after transplantation, it is important to predict the occurrence of aGVHD and prepare for it as well as to control the CsA dose.

Conclusions

We found that the incidence of aGVHD was significantly high regardless of elapsed time in weeks. In particular, we observed the highest incidence of aGVHD with the elapsed time of 7–13 days after we detected low CsA concentrations. Clinicians must pay careful attention to this elapsed time after they detect low CsA concentrations in order to prevent aGVHD.

Declarations

- *Ethics approval and consent to participate*

The study was approved by the Asan Medical Center Institutional Review Board (IRB number: 2017-0509). Informed consents were obtained from children's parents.

- *Consent for publication*

Not applicable.

- *Availability of data and material*

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

- *Competing interests*

The authors declare that they have no competing interests" in this section.

- *Funding*

Not applicable.

- *Authors' contributions*

EKC, JY, and HSG made substantial contributions to conception and design of study. EKC and JYK made acquisition and analysis of data. JY and HSG made an interpretation of data. EKC, JY, and HSG have been involved in drafting and revising the manuscript. All authors read and approved the final manuscript.

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