Original article

Risk factors for diabetes mellitus and impaired glucose tolerance following allogeneic hematopoietic stem cell transplantation in pediatric patients with hematological malignancies

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Abstract

Long-term surviving recipients of allogeneic hematopoietic stem cell transplantation (HSCT) often suffer from diabetes mellitus (DM). We sought to identify risk factors for the development of post-transplant DM and impaired glucose tolerance (IGT) in pediatric HSCT patients. Glucose tolerance statuses were evaluated in 22 patients aged 6.3–21.8 years who had received allogeneic HSCT between the ages of 0.8–13.5 years. Five patients were diagnosed as having type 2 DM, and treated with insulin or oral hypoglycemic agents. Five patients were included in the IGT group, and the remaining 12 children were in the normal glucose tolerance (NGT) group. The cumulative incidence of DM plus IGT was 11.6% at five years and 69.3% at 10 years. None of the patients were obese/overweight and none had a family history of DM. There were no significant differences in serum levels of leptin and adiponection between the DM+IGT and the NGT groups. An average preprandial glucose levels in the DM+IGT group were significantly higher than those in the NGT group from preparative conditioning to 60 days after HSCT. In multivariate analysis, an age of ≥ 6 years at the time of HSCT was significantly associated with the development of DM+IGT. Additionally, careful follow-up is necessary, even for NGT patients.

Introduction

Allogeneic hematopoietic stem cell transplantation (HSCT) is an effective treatment for childhood hematological malignancies [1]. Most long-term survivors of allogeneic HSCT, however, experience transplant-related sequelae, particularly endocrine disorders [2–3]. In recent years, patients who had undergone allogeneic HSCT during childhood or adolescence have been reported to develop diabetes mellitus (DM) [2–10]. In young adults, DM onset could worsen the long-term quality of life, because these patients might suffer from the early appearance of complications, such as cardiovascular disease. Taskinen et al. [5] found that the variables associated with hyperinsulinemia in bone marrow transplantation (BMT) patients were the time from transplantation, the presence of chronic graft-versus-host disease (GVHD), and hypogonadism. On the other hand, Hoffmeister et al. [4] reported that the risk factors for type 2 DM in pediatric HSCT (autologous, syngeneic, and allogeneic) survivors were a diagnosis of acute or chronic leukemia, race/ethnicity other than non-Hispanic white, family history of DM, and asparaginase toxicity. Armenian et al. [11] demonstrated that grade II–IV acute GVHD and total body irradiation (TBI) conditioning were associated with an increased risk of DM in autologous and allogeneic HSCT recipients (from infants to elderly patients). Accordingly, there are controversies regarding the risk factors associated with post-HSCT DM. In this study, we assessed glucose tolerance in patients who underwent pediatric allogeneic HSCT and attempted to identify the risk factors associated with abnormal glucose metabolism.

Patients and methods

Patients

We retrospectively examined the medical records of 22 patients who underwent allogeneic HSCT between 1996 and 2010 at the Department of Pediatrics, Shinshu University Hospital. The criteria for participation in this study included the following: a period of ≥2 years after allogeneic HSCT; ongoing follow-up at the Department of Pediatrics, Shinshu University Hospital; and the performance of glucose tolerance statuses of patients. An oral glucose tolerance test (OGTT) was performed on 21 patients. Patient demographics and details are provided in Tables 1 and 2. The underlying diseases were acute lymphoblastic leukemia (n = 7), acute myeloid leukemia (n = 11), mixed lineage leukemia (n = 2), juvenile myelomonocytic leukemia (n = 1), and blastic NK cell lymphoma (n = 1). The median ages at diagnosis and at the first allogeneic HSCT were 5.1 years (range, 0.4–10.5 years), and 7.4 years (range, 0.8–13.5 years), respectively. The median age at the glucose tolerance status evaluation was 14.1 years

(range, 6.3–21.8 years), and the median duration from the first allogeneic HSCT to the evaluation was 5.9 years (range, 2.0–14.7 years). No patients had asparaginase toxicity.

The study protocol was approved by the institutional review board of The Shinshu University School of Medicine, and written informed consent was obtained from the parents and the patients themselves if patient age was ≥16 years of age.

Transplantation procedure

The conditioning regimen for each patient is shown in Table 2. Patient no. 10 underwent prophylactic cranial irradiation at an 18-Gy dose 6 years before allogeneic HSCT. Twenty-one patients received TBI-based preparative conditioning for a first allogeneic HSCT (16 patients, 8 Gy; 1 patient, 10 Gy; and 4 patients, 12 Gy). Patient no. 7 received a busulfan-based conditioning regimen. Patient no. 22 received a 7-Gy craniospinal irradiation and a 3-Gy whole-brain irradiation 5 months after undergoing 8-Gy TBI. Three of the 22 patients underwent ≥2 allogeneic HSCTs. For the second and third HSCTs in patients no. 5 and 21, we performed chemotherapy alone as preparative conditioning. Chemotherapy combined with local 20-Gy irradiation (orbital cavity) was used as conditioning before the second allogeneic HSCT in patient no. 19.

Donors

Human leukocyte antigens (HLA) for A, B, and DRB1 in donors as well as recipients were determined by high-resolution DNA typing. The stem cell source and HLA disparity in each HSCT is described in detail in Table 2. For 3 patients (no. 5, 19, and 21) who underwent multiple allogeneic HSCTs, the donors were relatives of the patients. In the remaining 19 patients, the stem cell sources were relative BM (n = 6), non-relative BM (n = 3), relative peripheral blood (PB, n = 4), and non-relative cord blood (CB, n = 6).

Prophylaxis for GVHD

As a prophylactic treatment for GVHD, cyclosporin A and short-term methotrexate (sMTX) was used in 6 cases, tacrolimus+sMTX+methylprednisolone (mPSL) in 18 cases, and sMTX alone in 2 cases.

Anthropometric measurements

After measuring the body weight and height, the body mass index (BMI) was calculated by dividing the body weight in kilograms by the squared body height in meters (kg/m^2). For patients aged <17.5 years, the BMI percentile was determined with the calculation software invented by a joint committee on growth reference values, The Japanese Society for Pediatric Endocrinology, and the Japanese Association for Human Auxology

(http://jspe.umin.jp/taikakushisuv1.xlsx). Patients with a BMI of ≥90th percentile according to age and gender were classified as overweight or obese. For patients aged ≥17.5 years, patients with BMIs of ≥25 kg/m^2 and ≥30 kg/m^2 were classified as overweight and obese, respectively, according to the definitions from the World Health Organization [12].

Evaluation of glucose tolerance status

The insulin, hemoglobin A1c (HbA1c), and fasting plasma glucose levels were measured. During the acute phase after allogeneic HSCT, the blood glucose levels were measured daily or once every few days, early in the morning before meals. We then counted the number of times that the preprandial blood glucose levels exceeded 150 mg/dL from 30 days before the start of preparative conditioning to 60 days after HSCT in each patient, because the shortest length of hospital stay was 62 days after HSCT.

The HbA1c levels were expressed as National Glycohemoglobin Standardization Program equivalent values (%) and were calculated with the formula HbA1c = HbA1c (Japanese Diabetes Society) (%) + 0.4% [13].

The glucose tolerance statuses of 21 patients were evaluated using the OGTT. After an overnight fast, 1.75 g/kg (maximum of 75 g) of glucose were orally administered. The plasma glucose and insulin levels were measured in blood samples that were collected before and at 15, 30, 60, 90, and 120 min after the glucose loading. OGTT was performed once in 17 patients, and repeated OGTT was done in 4 patients.

The definitions of DM, impaired fasting glucose, and impaired glucose tolerance (IGT) were formulated on the basis of the diagnostic criteria established in 2010 by the American Diabetes Association [14]. Briefly, DM was defined as an HbA1c of ≥6.5%, a fasting plasma glucose level of ≥126 mg/dL, a plasma glucose level of ≥200 mg/dL at 2 h after an OGTT, or classic symptoms of hyperglycemia/hyperglycemic crisis with a random plasma glucose level of ≥200 mg/dL. Impaired fasting glucose was defined as a fasting plasma glucose level of 100–125 mg/dL. IGT was defined as a plasma glucose level of 140–199 mg/dL at 2 h after an OGTT. A group of patients who did not satisfy the criteria for DM, impaired fasting glucose, and IGT was defined as the normal glucose tolerance (NGT) group.

Insulin secretion, insulin resistance, and the sensitivity index were calculated as follows: the insulinogenic index = $(Ins_{30} - Ins_0)/(Glu_{30} - Glu_0)$, where Ins_y and Glu_y represent values at time y min during OGTT [15–16]; the homeostasis model assessment of insulin resistance (HOMA-IR) = $Glu_0 \times Ins_0/405$ [15, 17]; and the Matsuda insulin sensitivity index (ISI) =$10{,}000/(Glu_0 \times Ins_0 \times Glu_{120} \times Ins_{120})^{0.5}$. [15, 18–19] HOMA-IR values >2.6 were considered

diagnostic of insulin resistance [20].

Measurement of serum adipocytokines

The serum leptin levels were measured with a double-antibody radioimmunoassay. The adiponectin levels were measured with a latex turbidimetric immunoassay (SRL Inc., Tokyo, Japan).

Statistical methods

PASW Statistics, Version 18.0 (SPSS, Inc., Chicago, IL, USA) was used for the statistical analyses. The Mann–Whitney test or unpaired t-test was used to compare clinical parameters between the DM+IGT and NGT groups, as well as between the IGT group and NGT groups. The cumulative incidence of DM+IGT from allogeneic HSCT to the diagnosis day of DM or IGT was estimated according to the Kaplan-Meier method. To determine factors predictive of the development of DM+IGT, the patients' parameters were analyzed with the chi-squared test or Fisher's exact test. Then, the parameters displaying $p < 0.1$ in the univariate analysis were applied to logistic regression model. Statistical significance was defined as $p < 0.05$.

Results

Glucose tolerance statuses of 22 patients who underwent pediatric allogeneic HSCT

The fasting blood glucose levels at the onset of hematological malignancies were within the normal range in all 22 patients. As presented in Table 3, 5 patients were diagnosed with type 2 DM (DM group), on the basis of the diagnostic criteria by the American Diabetes Association [the fasting plasma glucose levels (≥126 mg/dL) and HbA1c levels (≥6.5 %)], upon evaluation at 4–12 years after allogeneic HSCT. According to the OGTT results, 4 patients had plasma glucose levels >200 mg/dL at 2 h after glucose loading. In patient no. 3, she was classified into the DM group by the third OGTT. Patient no. 4 was treated with insulin, whereas the other 4 patients were treated with oral hypoglycemic agents. In cases no. 6–10, the plasma glucose levels at 2 h after glucose loading were ≥140 mg/dL, although the fasting plasma glucose levels were <100 mg/dL and the HbA1c levels were <6.5%. In patient no. 8, he was classified into the IGT group from the second OGTT. Accordingly, these 5 patients were included in the IGT group. Patients no. 11–22 had fasting plasma glucose levels <100 mg/dL, HbA1c levels <6.5%, and normal responses to OGTT and were included in the NGT group. In patient no. 19, he showed IGT on the first OGTT during treatment with prednisolone, but displayed normal responses to the second and third OGTT without the steroid administration. Five patients with type 2 DM described above satisfied the diagnostic criteria by the Japan

Diabetes Society (http://www.jds.or.jp/modules/en/index.php?content_id=1). Five patients classified as the IGT group and 12 patients classified as the NGT group corresponded to borderline type and normal type, respectively, according to the Japanese criteria. The cumulative incidence of DM+IGT was 11.6% at 5 years and 69.3% at 10 years after HSCT.

All 5 patients in the DM group had a HOMA-IR of >2.6, and Matsuda ISI levels of 4 evaluated patients ranged from 1.7 to 2.9. The HOMA-IR levels of patients no. 7, 14, and 15 were 3.4, 2.7, and 2.8, respectively.

Seventeen patients were <17.5 years when the glucose tolerance statuses were evaluated. For these patients, the mean BMI percentile according to age and gender was 28.3. The remaining 5 patients were >17.5 years, and their mean BMI was 19.6. Thus, none of the patients in our study were obese or overweight.

Comparison of clinical findings between the DM+IGT group and NGT group

There were significant differences between the DM+IGT group and NGT group with regard to the fasting plasma glucose levels (122.7 ± 46.2 vs. 89.3 ± 5.2, $p < 0.001$), HbA1c levels (7.2 ± 2.1 vs. 5.5 ± 0.4, $p = 0.029$), fasting insulin levels (9.9 ± 3.1 vs. 6.3 ± 3.6, $p = 0.018$), HOMA-IR (3.0 ± 1.3 vs. 1.4 ± 0.8, $p = 0.002$), and Matsuda ISI (3.1 ± 1.1 vs. 8.9 ± 5.6, $p = 0.001$). Nevertheless, the insulinogenic index did not differ between the 2 groups (0.8 ± 0.6 vs. 0.8 ± 0.4, $p = 0.815$). The fasting plasma glucose levels and Matsuda ISI of the IGT group significantly differed from those of the NGT group (95.4 ± 1.7 vs. 89.3 ± 5.2, $p = 0.002$, and 3.3 ± 1.3 vs. 8.9 ± 5.6, $p = 0.006$, respectively).

The median age at diagnosis of hematological malignancies did not differ between the DM+IGT group and NGT group, with values of 5.8 years (range, 2.6–10.5 years) vs. 4.0 years (range, 0.5–9.9 years), respectively. The median age at first allogeneic HSCT was higher in the DM+IGT group than in the NGT group, with values of 9.1 years (range, 5.0–13.6 years) vs. 5.3 years (range, 0.8–12.6 years; $p = 0.047$), respectively. In addition, the median age at the glucose tolerance status evaluation was substantially higher in the DM+IGT group than in the NGT group, with values of 16.1 years (range, 9.6–21.9 years) vs. 9.7 years (range, 6.3–18.3 years; $p = 0.001$), respectively. The median interval between allo-HSCT and glucose tolerance status assessment study was also longer in the DM+IGT group than in the NGT group, with values of 78 months (range, 32–176 months) vs. 60 months (range, 24–90 months; $p = 0.031$), respectively.

We then compared the preprandial blood glucose levels in the morning from 30 days before the start of preparative conditioning to 60 days after HSCT between the DM+IGT group and NGT group. There was no

difference in the frequencies of blood sampling for 30 days before the start of preparative conditioning (10.6 ± 4.4 times in the DM+IGT group vs. 10.4 ± 4.1 times in the NGT group, p=0.923) and in the frequencies of blood sampling from the start of preparative conditioning to 60 days after HSCT (47.9 ± 14.6 times in the DM+IGT group vs. 51.3 ± 9.9 times in the NGT group, p=0.505). The prepandial blood glucose levels increased after the start of preparative conditioning in the DM+IGT group, when compared with the values obtained before preparative conditioning (101.4 ± 13.1 mg/dL vs. 117.8 ± 15.9 mg/dL, p<0.001), but not in the NGT group (95.4 ± 11.9 mg/dL vs. 98.8 ± 13.8 mg/dL, p=0.522). The mean prepandial blood glucose levels from the start of preparative conditioning to 60 days in the DM+IGT group were significantly higher than those in the NGT group (p=0.007), while there was no substantial difference in the values before the preparative conditioning between the two groups. Among 10 patients in the DM+IGT group, 7 patients developed hyperglycemia (≥150 mg/dL) 4 to 32 times, and one patient showed hyperglycemia twice. In the NGT group, 3 of 12 patients developed hyperglycemia (≥150 mg/dL) 4 to 12 times, and 4 patients showed hyperglycemia once to three times. Hyperglycemic episodes occurred sporadically or successively in the both groups.

Risk factors for post-transplant DM+IGT

To examine whether patients who would develop DM or IGT could be predicted before and soon after allogeneic HSCT, we first performed univariate analysis of the patients' pre-transplant information and clinical data after HSCT (Table 4) When the patients were separated into younger (<6 years of age) and older groups (≥6 years of age) at the time of allogeneic HSCT, DM+IGT occurred more frequently in the older group than in the younger group (p = 0.026). Patients with ≥4 times of hyperglycemia during acute phase of HSCT were more frequently observed in the DM+IGT group than in the NGT group (p=0.046). Multivariate analysis showed ≥6 years of age at the time of HSCT as the only significant risk factor for the development of DM or IGT (p=0.040). Repeated hyperglycemia was marginally associated with the occurrence of DM+IGT (p=0.056, Table 5).

On the other hand, there were no differences with regard to family history of DM and underlying diseases between the DM+IGT group and NGT group. Two or three allele-mismatched or unrelated HSCT did not influence DM+IGT development. Grade II–IV acute GVHD, chronic GVHD, tacrolimus use for GVHD prophylaxis or treatment, and corticosteroid use over 4 weeks after allogeneic HSCT all failed to influence DM+IGT development (Table 4).

Relationship between adipocytokines and DM+IGT development

The fasting/early morning serum leptin and adiponectin levels were measured in 19 cases. The leptin levels in the DM+IGT and NGT groups were 14.0 ± 11.7 ng/mL and 5.5 ± 3.0 ng/mL, respectively ($p = 0.106$), whereas the adiponectin levels were 8.3 ± 6.2 μg/mL and 11.5 ± 3.6 μg/mL, respectively ($p = 0.180$).

Discussion

In the present study, the cumulative incidence of DM+IGT in 22 allogeneic HSCT survivors was 11.6% at 5 years and 69.3% at 10 years. Among these patients, 21 received TBI (≥8 Gy)-based preparative regimens. Similar results were reported by Taskinen et al. [5], that 12 (52%) of the 23 children who underwent BMT developed IR (DM in 4 and IGT in 6). The authors of that study used TBI (10–12 Gy) as conditioning for 18 patients. Neville et al. [21] described that TBI conditioning increased the risk of hyperinsulinemia/IGT/DM development, whereas busulfan was protective. Therefore, TBI regimens appear to frequently cause post-transplant insulin resistance.

DM causes various types of chronic complications such as cardiovascular disease, which particularly worsens patients' quality of life. Therefore, we attempted to identify the risk factors to promote the prevention of and/or early intervention for post-transplant DM and IGT. The multivariate analysis revealed ≥6 years of age at the time of HSCT as only a significant risk for post-transplant DM+IGT. The more frequent occurrence of post-HSCT abnormal glucose metabolism after HSCT in the older patients might be related to the evidence that insulin resistance increases during adolescence [26–27]. A large part of patients in the DM+IGT group were older than 15 years at the time of the study, whereas only 2 of 12 patients in the NGT group were older than 15 years. Thus, we cannot deny a possibility that the older age at the time of HSCT in the DM+IGT group reflected the older age at the time of the study.

In the current study, an average of preprandial blood glucose levels early in the morning during acute phase of HSCT was significantly higher in the DM+IGT group than in the NGT group, although repeated hyperglycemia marginally associated with the development of DM+IGT according to multivariate analysis. Seven of 10 patients in the DM+IGT group experienced ≥4 episodes of hyperglycemia up to 60 days after HSCT. Sheean et al. [22–23] described that hyperglycemia occurs frequently during the acute post-transplant period because of intravenous hyperalimetation, glucocorticoid and immunosuppressant administration for GVHD, and infection. Accordingly, frequent hyperglycemia during HSCT might affect a patient's insulin resistance and insulin secretion capacity. On the basis of the results of 1,175 adult HSCT recipients as reported by Hammer et al. [24], 93% subjects experienced

hyperglycemia (blood glucose level ≥150 mg/dL) at least once during the acute phase after HSCT. However, most of the blood glucose abnormalities were transient. Even when the patients developed steroid-induced DM, hyperglycemia was generally improved by a decreased or discontinued corticosteroid dosage. Therefore, a wait-and-see approach was adopted in most cases. Fuji et al. [25] demonstrated that intensive glucose control reduced the incidences of infectious disease and organ dysfunction after allogeneic HSCT. A prospective study will be required to elucidate whether post-transplant DM+IGT can be alleviated with intensive glycemic control during the acute post-transplant period.

In contrast to previous reports regarding the general population, none of our HSCT survivors who developed DM+IGT were obese or overweight. These results are agree with those described by Taskinen et al. [5] and Hoffmeister et al. [4]. Adipocytokines such as leptin and adiponectin, which are produced and secreted by adipocytes, have been shown to be involved in insulin resistance development in obese subjects [28]. An increase in visceral fat causes a decrease in the blood adiponectin level, thus leading to insulin resistance [29]. By acting on receptors in the hypothalamus, leptin exerts a strong inhibitory effect on food intake and an enhancing effect on energy consumption; however, obese patients exhibit leptin resistance [30–31]. Annaloro et al. [32] reported that adult patients who developed metabolic syndrome after undergoing HSCT had lower serum adiponectin levels and elevated serum leptin levels. On the other hand, there were no significant differences in the adiponectin and leptin levels between the DM+IGT and NGT groups in our study. Taken together, these findings suggest that factors other than dysregulated adipocytokine production might be involved in the development of post-transplant DM+IGT. Nevertheless, we cannot exclude the possibility of abdominal obesity in the DM+IGT group.

Some patients in the NGT group showed elevated HOMA-IR values. In addition, patients in the NGT group were younger at the time of HSCT than those in the DM+IGT group. While a majority of patients in the DM+IGT group were in the adolescence or older at the time of the study, more than half of the NGT group were before puberty. Therefore, it is possible that the NGT group will develop IGT or DM later. Careful follow-up is necessary, even for NGT patients.

Conflict of Interest

We declare that we have no conflict of interest.

References

1. Horibe K, Saito AM, Takimoto T, Tsuchida M, Manabe A, Shima M, et al. Incidence and survival rates of hematological malignancies in Japanese children and adolescents (2006-2010): based on registry data from the Japanese Society of Pediatric Hematology. Int J Hematol. 2013;98:74-88.
2. Cohen A, Békássy AN, Gaiero A, Faraci M, Zecca S, Tichelli A, et al. Endocrinological late complications after hematopoietic SCT in children. Bone Marrow Transplant. 2008;41:S43-8.
3. Ishida Y, Honda M, Ozono S, Okamura J, Asami K, Maeda N, et al. Late effects and quality of life of childhood cancer survivors: part 1. Impact of stem cell transplantation. Int J Hematol. 2010;91:865-76.
4. Hoffmeister PA, Storer BE, Sanders JE. Diabetes mellitus in long-term survivors of pediatric hematopoietic cell transplantation. J Pediatr Hematol Oncol. 2004;26:81-90.
5. Taskinen M, Saarinen-Pihkala UM, Hovi L, Lipsanen-Nyman M. Impaired glucose tolerance and dyslipidaemia as late effects after bone-marrow transplantation in childhood. Lancet. 2000;356:993-7.
6. Bonanomi S, Gaiero A, Masera N, Rovelli A, Uderzo C, Fichera G, et al. Distinctive characteristics of diabetes mellitus after hematopoietic cell transplantation during childhood. Pediatr Transplant. 2006;10:461-5.
7. Traggiai C, Stanhope R, Nussey S, Leiper AD. Diabetes mellitus after bone marrow transplantation during childhood. Med Pediatr Oncol. 2003;40:128-9.
8. Shalitin S, Phillip M, Stein J, Goshen Y, Carmi D, Yaniv I. Endocrine dysfunction and parameters of the metabolic syndrome after bone marrow transplantation during childhood and adolescence. Bone Marrow Transplant. 2006;37:1109-17.
9. Tahrani AA, Cramp C, Moulik P. The development of non-insulin-dependent diabetes after total body irradiation and bone marrow transplantation in adolescence: a case report and literature review. Pediatr Diabetes. 2006;7:173-5.
10. Majhail NS, Challa TR, Mulrooney DA, Baker KS, Burns LJ. Hypertension and Diabetes Mellitus in Adult and Pediatric Survivors of Allogeneic Hematopoietic Cell Transplantation. Biol Blood Marrow Transplant. 2009;15:1100-07.
11. Armenian SH, Sun CL, Vase T, Ness KK, Blum E, Francisco L, et al. Cardiovascular risk factors in hematopoietic cell transplantation survivors: role in development of subsequent cardiovascular disease. Blood. 2012;120: 4505-12.

12. WHO. Obesity: preventing and managing the global epidemic. Report of a WHO Consultation. World Health Organ Tech Rep Ser. 2000;894:i-xii, 1-253.
13. Kashiwagi A, Kasuga M, Araki E, Oka Y, Hanafusa T, Ito H, et al. International clinical harmonization of glycated hemoglobin in Japan: From Japan Diabetes Society to National Glycohemoglobin Standardization Program values. J Diabetes Investig. 2012;3:8-10.
14. American Diabetes Association. Standards of medical care in diabetes-2010. Diabetes Care. 2010;33:S11-61.
15. Yeckel CW, Weiss R, Dziura J, Taksali SE, Dufour S, Burgert TS, et al. Validation of Insulin Sensitivity Indices from Oral Glucose Tolerance Test Parameters in Obese Children and Adolescents. J Clin Endocrinol Metab. 2004;89:1096-101.
16. Phillips DI, Clark PM, Hales CN, Osmond C. Understanding oral glucose tolerance: comparison of glucose or insulin measurements during the oral glucose tolerance test with specific measurements of insulin resistance and insulin secretion. Diabet Med. 1994;11:286-92.
17. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 1985;28:412-9.
18. Matsuda M, DeFronzo RA. Insulin sensitivity indices obtained from oral glucose tolerance testing: comparison with the euglycemic insulin clamp. Diabetes Care. 1999;22:1462-70.
19. DeFronzo RA, Matsuda M. Reduced time points to calculate the composite index. Diabetes Care. 2010;33:e93.
20. Ascaso JF, Pardo S, Real JT, Lorente RI, Priego A, Carmena R. Diagnosing insulin resistance by simple quantitative methods in subjects with normal glucose metabolism. Diabetes Care. 2003;12:3320-5.
21. Neville KA, Cohn RJ, Steinbeck KS, Johnston K, Walker JL. Hyperinsulinemia, impaired glucose tolerance, and diabetes mellitus in survivors of childhood cancer: prevalence and risk factors. J Clin Endocrinol Metab. 2006;91:4401-7.
22. Sheean PM, Freels SA, Helton WS, Braunschweig CA. Adverse clinical consequences of hyperglycemia from total parenteral nutrition exposure during hematopoietic stem cell transplantation. Biol Blood Marrow Transplant. 2006;12:656-64.
23. Sheean PM, Braunschweig C, Rich E. The incidence of hyperglycemia in hematopoietic stem cell transplant recipients receiving total parenteral nutrition: a pilot study. J Am Diet Assoc. 2004;104:1352-60.

24. Hammer MJ, Casper C, Gooley TA. The contribution of malglycemia to mortality among allogeneic hematopoietic cell transplant recipients. Biol Blood Marrow Transplant. 2009;15:344-51.
25. Fuji S, Kim SW, Mori S, Kamiya S, Yoshimura K, Yokoyama H, et al. Intensive glucose control after allogeneic hematopoietic stem cell transplantation: a retrospective matched-cohort study. Bone Marrow Transplant. 2009;44:105-11.
26. Levy-Marchal C, Arslanian S, Cutfield W, Sinaiko A, Druet C, Marcovecchio ML, et al. Insulin Resistance in Children Consensus Conference Group Insulin Resistance in Children: Consensus, Perspective, and Future Directions. J Clin Endocrinol Metab. 2010;95:5189-98.
27. Goran MI, Gower BA. Longitudinal study on puberal insulin resistance. Diabetes 2001: 50: 2444-2450.
28. Yu YH, Ginsberg HN. Adipocyte signaling and lipid homeostasis: sequelae of insulin-resistant adipose tissue. Circ Res. 2005;96:1042-52.
29. Kadowaki T, Hara K, Yamauchi T, Terauchi Y, Tobe K, Nagai R. Molecular mechanism of insulin resistance and obesity. Exp Biol Med. 2003;228:1111-7.
30. Friedman JM, Halaas JL. Leptin and the regulation of body weight in mammals. Nature. 1998;395:763-70.
31. Bjørbaek C, Kahn BB. Leptin signaling in the central nervous system and the periphery. Recent Prog Horm Res. 2004;59:305-31.
32. Annaloro C, Usardi P, Airaghi L, Giunta V, Forti S, Orsatti A, et al. Prevalence of metabolic syndrome in long-term survivors of hematopoietic stem cell transplantation. Bone Marrow Transplant. 2008;41:797-804.

Table 1 Characteristics of 22 allogeneic HSCT survivors in whom glucose metabolism was assessed

Median age at diagnosis	5.1 (0.4–10.5) years
Median age at 1st HSCT	7.4 (0.8–13.5) years
Median interval between the initial diagnosis and 1st HSCT	9.1 (3–92) months
Median age at evaluation of glucose tolerance status	14.1 (6.3–21.8) years
Median interval between 1st HSCT and the study for glucose tolerance status	5.9 (2.0–14.7) year
Gender	
Male	7
Female	15
Underlying diseases	
Acute lymphoblastic leukemia	7
Acute myeloid leukemia	11
Mix lineage leukemia	2
Juvenile myelomonocytic leukemia	1
Blastic NK lymphoma	1
Number of times of allogeneic HSCT	
Once	19
Twice	2
Thrice	1
Donor source	
Bone marrow	13
Related	10
Unrelated	3
Peripheral blood (related)	7
Cord blood (unrelated)	6
Conditioning regimen for 1st HSCT	
TBI containing <10 Gy	16
TBI containing ≥10 Gy	5
No TBI	1
GVHD prophylaxis	
FK ± sMTX ± mPSL	18
CyA ± sMTX	6
sMTX	2
Overweight at the time of this study	0

* Overweight is defined as a body mass index (BMI) of ≥90th percentile for age and gender for patients aged <17.5 years and a BMI ≥25 for patients aged > 17.5 years

CyA, cyclosporin A; FK, tacrolimus; GVHD, graft-versus-host disease; HSCT, hematopoietic stem cell transplantation; mPSL, methylprednisolone; sMTX, short-term methotrexate; TBI, total body irradiation

Table 2 Characteristics of the DM, IGT, and NGT groups

Patient no.	Sex	Disease (age at onset)	Age at 1st HSCT	Donor/stem cell source	HLA disparity	Conditioning regimen	GVHD prophylaxis	aGVHD	cGVHD
DM group									
1	M	AML (3y9m)	5y	S/PB	none	TBI(12 Gy)+CY+VP16	sMTX	III	no
2	F	Mixed lineage Leukemia (5y2m)	7y7m	S/PB	none	TBI (10 Gy)+VP16+MEL	sMTX	II	skin
3	F	AML (8y1m)	9y1m	M/BM	2	TBI(8 Gy)+CY+FLU	sMTX+mPSL+FK	III	skin
4	F	AML (8y8m)	9y7m	M/PB	none	TBI(12 Gy)+CY+VP16+ATG	sMTX+CyA	no	skin
5	F	ALL (10y5m)	10y10m	M/BM M/PB S/BM	1st HSCT, none 2nd HSCT, none 3rd HSCT, 1	TBI(8 Gy)+CY+FLU CY+FLU+IDA+CA CY+FLU+BU	sMTX+FK sMTX+CyA sMTX+FK	no	no
IGT group									
6	F	Blastic NK lymphoma (5y10m)	6y10m	F/BM	2	TBI(12 Gy)+CY+FLU	sMTX+FK	I	no
7	F	ALL (2y7m)	7y2m	S/PB	none	BU+MEL	sMTX+CyA	I	skin lung
8	M	ALL (5y3m)	9y2m	U/BM	none	TBI(8 Gy)+CY+FLU	sMTX+FK	III	skin
9	M	AML (8y3m)	9y11m	S/BM	none	TBI(8 Gy)+CY+FLU	sMTX+CyA	no	no
10	F	ALL (5y10m)	13y6m	U/BM	1	TBI(8 Gy)+CY+FLU	sMTX+mPSL+FK	no	skin
NGT group									
11	F	JMML (0y5m)	9m	U/CB	1	TBI(8 Gy)+CY+FLU	mPSL+ FK	no	no
12	F	AML (0y11m)	1y4m	U/BM	none	TBI(8 Gy)+CY+FLU	sMTX+FK	II	no
13	F	AML (3y)	3y9m	U/CB	1	TBI(8 Gy)+CY+FLU	mPSL+FK	I	no
14	F	AML (4y4m)	4y11m	S/BM	none	TBI(8 Gy)+CY+FLU	sMTX+ CyA	I	no
15	F	ALL (3y8m)	5y7m	M/BM	1	TBI(8 Gy)+CY+FLU	mPSL+sMTX+FK	II	no
16	M	AML (5y1m)	7y7m	U/CB	1	TBI(8 Gy)+CY+FLU	mPSL+ FK	II	no
17	F	AML (9y11m)	12y7m	M/BM	3	TBI(8 Gy)+CY+FLU	mPSL+sMTX+FK	III	skin
18	M	Mixed lineage leukemia (3y4m)	3y10m	U/CB	1	TBI(8 Gy)+CY+FLU	mPSL+FK	III	skin
19	M	AML (2y7m)	3y5m	M/BM M/PB	1st HSCT, 3 2nd HSCT, 3	TBI(8 Gy)+CY+FLU LI(20 Gy)+CY+FLU+IDA	sMTX+mPSL+FK sMTX+mPSL+FK	I III	skin skin
20	M	ALL (4y5m)	6y9m	U/CB	none	TBI(12 Gy)+CY+FLU	mPSL+FK	I	skin
21	F	AML (9y6m)	10y1m	F/BM F/PB	1st HSCT, none 2nd HSCT, none	TBI(8 Gy)+CY+FLU CY+FLU+IDA	sMTX+FK CyA	no	no
22	F	ALL (8y3m)	10y11m	U/CB	1	TBI(8 Gy)+CY+FLU	mPSL+FK	III	skin

aGVHD, acute GVHD; ALL, acute lymphoblastic leukemia; AML, acute myeloid leukemia; ATG, anti-thymocyte/lymphocyte globulin; BM, bone marrow; BU, busulfan; CA, cytarabine; CB, cord blood; cGVHD, chronic GVHD; CY, cyclophosphamide; CyA, cyclosporin A; DM, diabetes mellitus; F, father; FK, tacrolimus; FLU, fludarabine; GVHD, graft-versus-host disease; HLA, human leukocyte antigen; HSCT, hematopoietic stem cell transplantation; IDA, indarubicin; IFG, impaired fasting glycemia; IGT, impaired glucose tolerance; JMML, juvenile myelomonocytic leukemia; LI, localized irradiation; MEL, melphalan; M, mother; mPSL, methylprednisolone; sMTX, short-term methotrexate; NGT, normal glucose tolerance; PB, peripheral blood; S, sibling; TBI, total body irradiation; U, unrelated donor; VP16, etoposide

Table 3 Glucose metabolism in the DM, IGT, and NGT groups

Patients no.	Age at the study	Interval after HSCT (years)	BMI	HbA1c (%)	Fasting plasma glucose (mg/dL)	Plasma glucose level at 120 min (mg/dL)	Fasting insulin (μU/mL)	Insulinogenic index	HOMA-IR	Matsuda ISI
DM group										
1*	16y5m	11.3	18.9 (22.4)	7.7	104	260	13.1	0.2	3.4	2.3
2*	19y8m	12.1	20.4	7.2	106	202	11.1	1.1	2.9	1.6
3*	15y2m	6.1	16.6 (2.7)	8.8	164	399	14.3	0.1	5.8	1.7
4*	18y2m	8.6	21.8	12	236	ND	7.1	ND	4.1	ND
5*	15y9m	4.8	20.6 (46.6)	7.8	140	297	8.6	0.1	3.0	2.9
IGT group										
6*	9y6m	2.7	15.3 (26.3)	5.8	94	142	7.8	0.6	1.8	5.7
7	21y10m	14.7	15.5	5.7	94	143	14.5	1.2	3.4	2.3
8*	15y2m	5.9	19.0 (33.3)	5.2	98	154	6.1	1.2	1.5	3.1
9	15y10m	5.8	22.2 (70.9)	5.8	96	143	8.7	1.5	2.1	3.2
10	20y5m	6.9	19.6	6	95	153	7.9	1.0	1.9	2.5
NGT group										
11*	8y4m	7.5	14.5 (20.2)	5.5	97	125	4.7	0.9	1.1	10.9
12	6y9m	5.4	15.9 (57.1)	5.6	80	110	3	0.6	0.6	14.0
13	8y3m	4.5	18.0 (80)	5.4	92	107	8.2	1.0	1.9	7.6
14	9y6m	4.6	15.7 (34.3)	5.8	86	111	12.5	1.2	2.7	5.3
15	12y8m	7.1	16.5 (13.3)	5.8	90	132	12.8	-0.2	2.8	2.7
16	10y8m	3.1	16.0 (28.8)	5.2	84	111	3	0.9	0.6	17.0
17*	18y4m	5.7	15.1	5.9	93	110	2.7	1.0	0.6	54.9
18*	6y3m	2.3	12.9 (1.0)	4.8	92	87	4.2	0.8	1.0	23.3
19	9y10m	6.3	15.6 (28.2)	6.1	95	129	4	0.8	0.9	6.5
20	9y	2.1	17.4 (68.7)	5.1	82	87	4.8	0.4	1.0	13.0
21	16y4m	6.2	16.9 (2.3)	5.8	87	115	8.4	1.6	1.8	5.8
22	12y11m	2.0	15.8 (5.2)	5.3	92	129	7.7	0.7	1.8	4.5

*Patients with ≥4 morning preprandial blood glucose levels of 150 mg/dL within 60 days after HSCT.

BMI values are expressed as kg/m^2. In patients aged <17.5 years, the percentile is shown in parentheses.

BMI, body mass index; DM, diabetes mellitus; HSCT, hematopoietic stem cell transplantation; HOMA-IR, homeostasis model assessment of insulin resistance; IGT, impaired glucose tolerance; ISI, insulin sensitivity index; ND, not done; NGT, normal glucose tolerance

Table 4 Effects of pre/post-transplant factors on DM and IGT development

	DM+IGT (n = 10)	NGT (n = 12)	*p*
Age at HSCT			
6 years or older	9	5	0.026
Younger than 6 years	1	7	
Sex			
Male	3	4	0.454
Female	7	8	
Family history of DM			
Positive	4	2	0.229
Negative	6	10	
Underlying disease			
Myeloid	4	7	0.392
others	6	5	
Donor			
Related	8	5	0.082
Unrelated	2	7	
HLA disparity			
No allele mismatched or 1 allele-mismatched	8	10	0.632
2 or 3 allele-mismatched	2	2	
Conditioning regimen			
TBI containing <10 Gy	5	11	0.080
TBI containing ≥10 Gy	4	1	
Plasma glucose level			
<150 or ≥150 mg/dL three times or less	3	9	0.046
≥150 mg/dL four times or more	7	3	
FK use for GVHD prophylaxis/treatment			
Yes	7	11	0.226
No	3	1	
Acute GVHD			
Grades II–IV	4	7	0.392
Grades 0–I	6	5	
Chronic GVHD			
Presence	4	7	0.392
Absence	6	5	
Corticosteroid use			
Less than 4 weeks	4	2	0.229
Over 4 weeks	6	10	

DM, diabetes mellitus; FK, tacrolimus; GVHD, graft-versus-host disease;HLA, human leukocyte antigen; HSCT, hematopoietic stem cell transplantation; IGT, impaired glucose tolerance;mPSL, methylprednisolone; NGT, normal glucose tolerance; sMTX, short-term methotrexate; TBI, total body irradiation

Table 5 Predictive factors for the development of DM and IGT

Variables	Unfavorable factors	Multivariate analysis	
		Odds (95% CI)	*p*
Age	≥6 years at HSCT	19.39 (1.14-329.15)	0.040
Hyperglycemic episodes	≥4 times up to 60 days after HSCT	10.94 (0.94-126.96)	0.056
Donor	Related	4.62 (0.43-49.34)	0.205
TBI	≥10 Gy	3.55 (0.18-71.81)	0.409

Abbreviations: CI, confidence interval; DM, diabetes mellitus; HSCT, hematopoietic stem cell transplantation; IGT, impaired glucose tolerance; TBI, total body irradiation