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Endocrinopathies, Bone Health, and Insulin Resistance in Patients with Fanconi Anemia after Hematopoietic Cell Transplantation

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A B S T R A C T

A number of endocrinopathies have been described after hematopoietic cell transplantation (HCT), but data are limited in patients with Fanconi anemia (FA). We report several endocrine-based disorders in a cohort of 44 patients with FA after HCT compared with both 74 patients who received HCT for hematologic malignancies and with 275 healthy controls. Endocrinopathies assessed included hypothyroidism, hypogonadism, short stature, dyslipidemia, insulin resistance, abnormalities in body composition, and bone health. Most (86%) patients with FA had at least 1 endocrinopathy, with 11% having 3 or more. Hypothyroidism was seen in 57%, hypogonadism in 27%, short stature in 50%, and reduced total body and lumbar spine bone mineral density (BMD) (height adjusted Z-score < -1) in 57% and 21%, respectively. Vitamin D deficiency was seen in 71%. Short stature was associated with younger age at HCT and gonadal failure was associated with older age at HCT. Insulin resistance was associated with increased percent fat mass and increased android/gynoid ratio by dual energy X-ray absorptiometry. Hypothyroidism, short stature, and reduced total body BMD were more prevalent in patients with FA compared with patients with hematologic malignancies. We recommend an assessment before transplantation and close follow-up afterwards to ensure proper clinical management. Future studies should continue to explore the impact of HCT on endocrinopathies in FA patients.

INTRODUCTION

Outcomes after hematopoietic cell transplantation (HCT) for patients with Fanconi anemia (FA) have improved significantly in recent years [1,2]. Survival after alternative-donor HCT with a fludarabine-containing regimen in patients with FA without prior opportunistic infections or transfusions is 94% at 5 years [3]. Therefore, long-term complications need to be investigated, particularly in this population with an increased rate of endocrinopathies at baseline [4]. Data on endocrinopathies after HCT in patients with FA are limited, with most reports combining late effects among patients with a variety of malignant and nonmalignant diseases [5-11].

Our primary aim was to comprehensively evaluate the endocrine, bone health, and metabolic profile of patients with FA who underwent allogeneic HCT using a uniform total body irradiation−(TBI) containing conditioning regimen. Our secondary aims were to evaluate for associations of these endocrine deficits with patient characteristics and to compare these outcomes to both a healthy sibling control group (healthy control) and with patients who received HCT for a hematologic malignancy (cancer HCT). We hypothesized that in patients with FA, the following would be observed: (1) insulin resistance would be associated with increased central adiposity, increased percent fat mass, and

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increased triglycerides; and (2) the prevalence of endocrinopathies would be higher in patients who underwent HCT for FA compared with those who underwent HCT for hematologic malignancies because of an increased risk of endocrinopathies in patients with FA at baseline.

MATERIALS AND METHODS

The institutional review boards at the University of Minnesota and the Fred Hutchinson Cancer Research Center/Seattle Children's Hospital approved the study. All patients and/or guardians signed institutional review board–approved informed consent for HCT and data collection in accordance with the Declaration of Helsinki. Studies were registered at http:www.clinicaltrials.gov under NCT00352976 and NCT00920842.

Study Participants

FA HCT

Our case population included 44 patients with FA who underwent allogeneic donor HCT for severe marrow failure at less than 35 years of age between 2006 and 2013 at the University of Minnesota with at least 1 year of follow-up. All were treated on the same protocol, with a single dose of TBI 300 cGy with thymic shielding, cyclophosphamide of 10 mg/kg for 4 days, fludarabine of 35 mg/m^2 for 4 days, with or without antithymocyte globulin and methylprednisolone with mycophenolate mofetil and cyclosporine or sirolimus as graft-versus-host disease (GVHD) prophylaxis as previously reported [3]. Donor grafts were T cell depleted related (n = 3) and unrelated (n = 27) donor marrow, unrelated donor single cord blood (n = 12), and unrelated donor double cord blood (n = 2).

Cancer HCT

This control group comprised 74 patients who received myeloablative HCT for hematologic malignancies at the Fred Hutchinson Cancer Center and the University of Minnesota between 1975 and 2008, as previously reported [12]. Diseases included acute myeloid leukemia (n = 32), acute lymphoblastic leukemia (n = 20), myelodysplastic syndrome (n = 12), chronic myelogenous leukemia (n = 7), non-Hodgkin lymphoma (n = 2), and juvenile myelomonocytic leukemia (n = 1). Sixty patients received chemotherapy and TBI (median, 1320; range, 750 to 1575 cGy) and 14 received chemotherapy alone. Donor grafts were T cell replete related (n = 40) and unrelated (n = 17) donor marrow, related (n = 1) and unrelated (n = 13) donor cord blood, and related (n = 2) and unrelated (n = 1) donor peripheral blood stem cells.

Healthy control

This control group consisted of 275 healthy siblings of patients with cancer [12,13]. Controls were excluded if they had a history of malignancy, previous HCT, or known chronic illness, such as hypothyroidism.

Study Procedures

Clinical and laboratory data were systematically and prospectively collected annually after HCT. All data were obtained from patients with FA; all data except metabolic parameters and vitamin D levels were obtained from both control groups. Endocrinologists performed Tanner staging. For females, separate breast and pubic hair staging were performed and the higher of these was recorded as the patient's Tanner stage. For males, only pubic hair Tanner stage was recorded since males with FA can have small genitalia. Chemiluminescent immunoassays were used to measure 25-hydroxyvitamin D, estradiol, thyroid stimulating hormone (TSH), follicle stimulating hormone (FSH), and luteinizing hormone levels. Free thyroxine was measured by competitive immunoassay and total testosterone by liquid chromatography/tandem mass spectrometry. Dual energy X-ray absorptiometry (DXA) scans were used to measure bone mineral density (BMD). DXA measures included Z-scores for total body BMD (TBMD) and including head and posterior anterior lumbar spine BMD (LBMD) at L1 to L4. BMD Z-scores were adjusted for sex and age, using reference data based on healthy ambulatory subjects from the general population who were free from chronic diseases affecting bone and not taking bone-altering medications. BMD Z-scores were also height-adjusted at the total body and lumbar spine ($TBMD_{HAZ}$ and $LBMD_{HAZ}$, respectively), using a method described by Zemel et al. [14]. The android to gynoid ratio (A/G) ratio and percent fat mass were measured by DXA, as previously described [15]. To better characterize adiposity beyond body mass index (BMI), percent fat mass was used to organize patients into normal, moderate, and elevated categories by age and sex, as previously defined [16]. Glycemic abnormalities and measures of insulin resistance were evaluated by oral glucose tolerance testing in the FA HCT group. After an overnight fast, 1.75 g/kg (maximum of 75 grams) of oral glucose was ingested, and glucose and insulin were measured by chemiluminescent immunoassay at 0, 30, 60, 90, and 120 minutes. The Matsuda index was used to calculate insulin resistance and was deemed abnormal if < 4.5 [17-19]. *Hypothyroidism* was defined by treatment with thyroid hormone replacement at the time of evaluation, free T4 < .7 ng/dL, or TSH > 5.0 mU/L before 2014 and > 4.0 mU/L starting in July of 2014, because of a change in the normative reference data. *Hypogonadism* was defined by treatment with estrogen or FSH > 40 IU/L in a female > 10 years of age. In a male, *hypogonadism* was defined as treatment with testosterone, FSH > 18 IU/L, or luteinizing hormone > 10 and low testosterone in a male > 11 years of age. Gonadal hormone levels were obtained in all females > 10 years and males > 11 years old. *Short stature* was defined as a height Z-score of less than -2. *Vitamin D deficiency* was defined as a 25-hydroxyvitamin D level of less than 30 ug/L. *Reduced BMD* was defined as a height-adjusted Z-score < -1.

Statistical Analysis

Descriptive statistics were expressed as frequencies and percent or mean/median ± standard deviation (SD), and range, as appropriate. For data involving healthy controls or repeated measures, *P* values were generated from generalized estimating equations with robust standard errors in the form of linear (continuous variable), logistic (binary variable), or multinomial regression (categorical variable). Otherwise, *P* values were calculated from the 2-sample *t*-test or the Chi-square test. If the overall test for 3-group comparison showed significant, pairwise comparison was done. Matsuda index was transformed in natural log, and its association with the A/G ratio was examined using linear mixed model. All analyses were done using the SAS system (v. 9.3; SAS Institute, Cary, NC). *P* values were 2-sided with < .05 considered statistically significant. Bonferroni adjustment was applied for multiple comparisons.

RESULTS

Characteristics of Study Participants

There was no difference in gender distribution among the 3 groups (Table 1). There was no difference in age at HCT for the FA HCT and cancer HCT groups. The cancer HCT group had longer follow-up and were, thus, older and more sexually mature at the latest evaluation. Both FA HCT patients and cancer HCT patients were shorter than healthy controls ($P < .001$). A significantly higher proportion of cancer HCT patients developed acute GVHD (57% versus 11%, $P < .001$)

Table 1
Characteristics of Study Participants

Characteristic	A FA HCT n = 44	B Cancer HCT n = 74	C Healthy Control n = 275	Pairwise *P* Values		
				A versus B	A versus C	B versus C
Sex, male	24 (54.6)	46 (62.2)	150 (54.6)	NS	NS	NS
Age at HCT, mean (SD) range, yr	10.7 (7.2) 3.3-34.3	10.5 (6.0) .6-22.6		NS		
Years since HCT, mean (SD) range	3.1 (1.9) 1.0-8.1	11.1 (5.9) 2.7-26.9		**<.001**		
Age at last study, mean (SD) range, yr	13.8 (7.1) 5.9-36.3	21.6 (6.3) 11.0-36.5	15.6 (4.7) 9.0-36.4	**<.001**	NS	**<.001**
Height Z-score, mean (SD), range	−1.8 (1.1) −4.7-.3	−.6 (1.1) −3.1-1.9	.4 (1.0) −1.8-3.1	**<.001**	**<.001**	**<.001**
Tanner stage, median (SD), range	1.0 (1.4) 1-5	5.0 (1.3) 1-5	4.0 (1.4) 1-5	**<.001**	**<.001**	**<.001**

NS indicates not statistically significant.
Items in bold are statistically significant.
P value < .0167 is considered statistically significant after multiple comparison adjustment using Bonferroni method.