hyperthyroidism was diagnosed and 2 were being treated for chronic GvHD; one with Etanercept, Cyclosporine and Solumedrol and one with Orapred and Imuran. One patient was still on prophylactic Tacrolimus when hyperthyroidism was diagnosed 9 months after transplant. All patients required a form of ablative therapy. Two received radioactive iodine as treatment. Three patients received Methimazole alone. One patient failed Methimazole, received radioactive iodine and then underwent thyroidectomy revealing follicular carcinoma. One of the patients did not receive any therapy; he had Hoshimoto's thyroidits and went on to develop hypothyroidism. Another patient had a total thyroidectomy that revealed diffuse hyperplasia. All patients are surviving 1 to 9 years after diagnosis of thyroid disease.

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Neuroblastoma Stem Cells: Identifying Two New Classes of Drugs with in-Vitro Activity in Cell Populations with High Side-Population Profiles

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Background: Neuroblastoma (NB) is a pediatric tumor of neural crest origin. It is one of several small round blue cell tumors. Patients who have MYC-N amplified tumors that respond sub-optimally to initial therapies have poor outcomes with 30-40% long-term survival. These tumors may have a higher content of therapy resistant cancer stem cells that utilize autophagy as an enhanced survival mechanism. In the following study we identified a stem cell phenotype in an MYC-N amplified neuroblastoma cell line from ATCC (BE-2) and investigated the activity of two new classes of drugs (GLUT-1 inhibitors and bisquinolone antimalarials).
Methods: Flow cytometry was utilized to identify stem cell fractions within an unselected BE-2 population. Briefly, in side population analysis 1 x 10^6 cells were incubated with Hoescht IMDM staining media (1 mcg/ml) and incubated for 30 minutes with Hoechst 33342 dye. Cells were then kept on ice until flow cytometry analysis. Flow cytometry was performed using a BD FACSAria. Autophagy was measured using a flow cytometry based assay measuring membrane bound LC3 (Milipore Inc). Screening assays for drug activity used a 96 well platform with MTT to measure cytotoxicity of bisquinolone and GLUT-1 cytotoxicity. Neurospheres were grown in non-adherent plates to assess activity in this stem cell enriched screening assay.
Results: Flow cytometry demonstrated a high percent of side population cells on three separate analyses (11%, 38%, 28% of live cells). In comparison two murine breast cancer cells lines (Clone 66 and 4T1) and human mantle cell line (Granta) side population fractions were approximately 1% of live cells. A commercially available bisquinolone, hydroxychloroquine, demonstrated the ability to inhibit autophagy as measured by the flow cytometry assay. Using the two *in-vitro* screening assays described above, a number of bisquinolone antimalarials and GLUT-1 inhibitors were assessed for activity and three compounds were identified with high activity (Q2-15 antimalarial and STF-04 and STF-05 GLUT-1 inhibitor).
Conclusion: In the following study we describe a large cancer stem cell population as measured by side population assay in BE-2 human neuroblastoma cells. Bisquinolone antimalarial drugs demonstrated the ability to inhibit autophagy. Both bisquinolone antimalarials and GLUT-1 inhibitors demonstrated cytotoxic activity in an MTT assay and activity in a stem cell enriched neurosphere assay. These two classes of drugs have the potential to improve therapy in poor prognostic neuroblastoma and studies combining candidate compounds are ongoing both in cell culture and an NSG animal model.

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Prospective Echocardiographic Screening for Cardiac Dysfunction 100 Days after Transplant in Children and Young Adults after Stem Cell Transplant

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Background: Cardiac evaluation during first 100 days after stem cell transplant (SCT) is usually performed only if clinically indicated, however, patients with mild left ventricular (LV) dysfunction can be asymptomatic. Scheduled post-SCT screening at day 100 may identify patients with subacute cardiac toxicity identified as LV dysfunction.
Methods: We conducted a single center prospective study to screen for LV dysfunction after SCT in 100 consecutive patients. Patients received echocardiography screening prior to SCT and 100 days post-SCT. Patients were classified as having LV dysfunction if echocardiography met at least one of the

Table 1

	LV Dysfunction at day +100 (n=23)	No LV Dysfunction at day +100 (n=69)	p value
Male	14 (61%)	47 (68%)	0.61
Mean Age in Yrs (95% CI)	9.6 (6.3-12.9)	6.9 (5.5-8.2)	**0.068**
Diagnosis			0.36
Malignancy	12 (51%)	22 (32%)	
Immune deficiency	5 (21%)	29 (42%)	
Bone Marrow Failure	5 (21%)	15 (22%)	
Genetic/Metabolic	1 (7%)	2 (3%)	
Benign Hematology	0	1 (1%)	
Therapy / Conditioning			
Previous anthracycline	11 (48%)	18 (26%)	**0.070**
RIC	12 (52%)	42 (61%)	0.47
Myeloablative	11 (48%)	27 (39%)	
Donor			0.11
Autologous	7 (30%)	9 (13%)	
Allogeneic	16 (70%)	60 (87%)	
Source			**0.032**
Bone Marrow	11 (48%)	43 (62%)	
Cord	0	8 (12%)	
PBSC	12 (52%)	18 (26%)	
Type			0.14
Auto	7 (30%)	9 (13%)	
Related Donor	3 (13%)	8 (12%)	
Unrelated Donor	13 (57%)	52 (75%)	
Transplant Complications			
GVHD	5 (22%)	26 (38%)	0.21
Engraftment Syndrome	1 (4%)	11 (16%)	0.28
Adenovirus	4 (17%)	12 (17%)	1.00
CMV	3 (13%)	19 (28%)	0.26
EBV	7 (30%)	31 (45%)	0.33
Death at 1 year	5 (22%)	14 (20%)	1.00