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ORIGINAL REPORT

Antitumor Activity of Hu14.18-IL2 in Patients With Relapsed/Refractory Neuroblastoma: A Children's Oncology Group (COG) Phase II Study

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Terms in blue are defined in the glossary, found at the end of this article and online at www.jco.org.

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ABSTRACT

Purpose
The hu14.18-IL2 fusion protein consists of interleukin-2 molecularly linked to a humanized monoclonal antibody that recognizes the GD2 disialoganglioside expressed on neuroblastoma cells. This phase II study assessed the antitumor activity of hu14.18-IL2 in two strata of patients with recurrent or refractory neuroblastoma.

Patients and Methods
Hu14.18-IL2 was given intravenously (12 mg/m^2/daily) for 3 days every 4 weeks for patients with disease measurable by standard radiographic criteria (stratum 1) and for patients with disease evaluable only by [^{123}I]metaiodobenzylguanidine (MIBG) scintigraphy and/or bone marrow (BM) histology (stratum 2). Response was established by independent radiology review as well as BM histology and immunocytology, and durability was assessed by repeat evaluation after more than 3 weeks.

Results
Thirty-nine patients were enrolled (36 evaluable). No responses were seen in stratum 1 (n = 13). Of 23 evaluable patients in stratum 2, five patients (21.7%) responded; all had a complete response (CR) of 9, 13, 20, 30, and 35+ months duration. Grade 3 and 4 nonhematologic toxicities included capillary leak, hypoxia, pain, rash, allergic reaction, elevated transaminases, and hyperbilirubinemia. Two patients required dopamine for hypotension, and one patient required ventilatory support for hypoxia. Most toxicities were reversible within a few days of completing a treatment course and were expected based on phase I results.

Conclusion
Patients with disease evaluable only by MIBG and/or BM histology had a 21.7% CR rate to hu14.8-IL2, whereas patients with bulky disease did not respond. Hu14.18-IL2 warrants further testing in children with nonbulky high-risk neuroblastoma.

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INTRODUCTION

Most children with neuroblastoma present with metastatic disease and/or high-risk features.[1,2] Despite multimodal intensive induction and consolidation therapy that provides responses for approximately 80% of patients, fewer than 40% of patients with high-risk disease are cured.[2,3] The majority of responding patients eventually die from recurrent disease, indicating that they still harbor viable neuroblastoma after front-line therapy.

The GD2 disialoganglioside is expressed on most neuroblastomas and melanomas and weakly on peripheral nerves.[4-6] Clinical trials using murine (3F8 and 14.G2a) and chimeric (ch14.18) anti-GD2 monoclonal antibodies (mAbs) have shown controllable toxicity (including pain and fever), but rare antitumor effects against measurable disease.[7-11] Preclinical data suggest in vivo activity is mediated by antibody-dependent cell-mediated cytotoxicity (ADCC) and is most effective in the minimal residual disease setting.[12-15] ADCC may be enhanced by interleukin-2 (IL-2), which activates natural killer (NK) cells,[16,17] and by granulocyte-macrophage colony-stimulating factor (GM-CSF), which activates neutrophils and macrophages.[18] Clinical trials have administered anti-GD2 mAbs together with IL-2 and/or GM-CSF.[19-26] Recently a Children's

Oncology Group (COG) phase III trial in patients with high-risk neuroblastoma showed a 66% versus 46% ($P = .01$) advantage in event-free survival (EFS) and a 86% versus 75% ($P = .02$) advantage in overall survival (OS) using a regimen of ch14.18 plus GM-CSF plus IL-2 and isotretinoin versus isotretinoin alone.[27]

The hu14.18-IL2 fusion protein consists of the humanized 14.18 anti-GD2 mAb linked to IL-2.[28] Hu14.18-IL2 localizes to GD2-positive tumor cell surfaces via the mAb component. The IL-2 component binds to and activates both NK and T cells via their IL-2 receptors, whereas the Fc end triggers ADCC and complement-dependent cytotoxicity (Buhtoiarov et al, manuscript submitted for publication).[28-30] Hu14.18-IL2 has preclinical activity in neuroblastoma-bearing mice via NK-mediated effects, especially when there is a smaller tumor burden.[14,31] In mice hu14.18-IL2 has superior antitumor activity compared with ch14.18 mAb combined with IL-2.[13,32]

Phase I testing of hu14.18-IL2 demonstrated biologic activity, clinical tolerability, and a maximum-tolerated dose of 12 mg/m^2/d for 3 days.[33,34] Dose-limiting toxicities (DLT) included hypotension and allergic reactions.

The primary objective of this study was to determine the antitumor activity of hu14.18-IL2 in subjects with measurable disease and subjects with disease evaluable only by [^{123}I]metaiodobenzylguanidine (MIBG) scintigraphy and/or bone marrow (BM) histology.

PATIENTS AND METHODS

Eligibility

Patients with recurrent or refractory neuroblastoma (age, 12 months to 22 years) were eligible. Primary refractory disease (persistent tumor after front-line therapy) required a biopsy demonstrating viable tumor. There were no prior therapy limitations. Eligibility required organ function, performance status, recovery from prior therapy, and life expectancy standard for COG phase II trials. Patients with CNS disease were excluded, as were patients requiring immunosuppression. Institutional review board–approved informed consent (and assent when applicable) was obtained for all patients.

Study Design

This phase II, single-arm trial evaluated the activity of hu14.18-IL2 separately for two patient strata. Stratum 1 included patients with disease measurable by computed tomography and/or magnetic resonance imaging using standard radiographic criteria. Stratum 2 included patients with disease evaluable only by ^{123}I-MIBG scintigraphy and/or BM histology.

Hu14.18-IL2 (EMD 273063) was supplied collaboratively by the National Cancer Institute (Bethesda, MD) as well as EMD Pharmaceuticals (Durham, NC) and Merck KGaA (Darmstadt, Germany). Hu14.18-IL2 (12 mg/m^2/dose) was administered on an inpatient basis as a 4-hour intravenous infusion over 3 consecutive days. Patients received indomethacin (0.5 mg/kg/dose, every 6 hours). Treatment cycles were 28 days. Toxicities were graded by the National Cancer Institute Common Toxicity Criteria (v3.0). DLT was defined as any grade 3 or worse toxicity, with certain reversible exceptions identified in the phase I studies.[33,34] Treatment was held for DLT and restarted at 50% of the previous dose once toxicity resolved. Disease evaluations were done every two courses.[35] Treatment was continued for four courses in the absence of progressive disease or drug intolerance. Subsequent treatment could continue for two courses after reaching a complete response (CR).

Evaluation of Response

All patients who completed two or more courses of hu14.18-IL2 or who had an event (relapse or progressive disease) were evaluable for response. All responses were confirmed by independent radiology review and marrow immunocytology.

The International Neuroblastoma Response Criteria were used to define response.[36] For measurable disease, response was determined using the Response Evaluation Criteria in Solid Tumors (RECIST). Response for stratum 2 patients was determined as follows:

MIBG response. Patients graded locally with CR or partial response (PR) for MIBG were scored by central review using the Curie scale.[37] CR was defined by complete resolution of all MIBG-avid lesions.

BM response. For patients who entered with BM disease (neuroblastoma identified in the BM aspirate and/or biopsy by the local pathologist using standard histology), CR was defined as no tumor cells detectable by morphology and immunocytologic analysis on two subsequent bilateral BM aspirates/biopsies done $\geq$ 3 weeks apart. Progressive disease (PD) was defined as $\geq$ 25% tumor in the marrow and a doubling in the percentage of tumor. Stable disease (SD) was defined as persistence of disease that does not meet criteria for CR or PR. Patients who cleared morphologic tumor but still had immunocytochemistry-detectible tumor (sensitive to 1 tumor cell in 1×10^5 nucleated cells)[35] were classified as having SD.

Immunologic Monitoring

Absolute lymphocyte counts were determined at each institution pretreatment and on days 1, 3, 4, 8, and 15 of each course. Serum samples were obtained pretreatment, immediately after treatment on days 1 and 3, and on days 4 and 8 of each course. These were analyzed for hu14.18-IL2 levels, anti-hu14.18-IL2 antibody, and soluble IL-2 receptor (sIL2R).[38,39]

Statistical Considerations

The primary end point of this study was response. Responders were defined as evaluable patients who demonstrated a best overall response of CR, very good partial response, or PR. Using a one-stage rule, if four or more

Table 1. Patient Characteristics by Stratum

Characteristic	Stratum 1 (n = 15)* No.	%	Stratum 2 (n = 24)† No.	%	Total (n = 39) No.	%
Eligible patients	15	100	24	100	39	100
Patients evaluable for toxicity	14	93	24	100	38	97
Patients evaluable for response	13	87	23	96	36	92
Age at diagnosis, months						
< 18	0	0	0	0	0	0
≥ 18	15	100	24	100	39	100
INSS stage						
1, 2, 3, 4s	0	0	2	8	2	5
4	11	73	15	63	26	67
Unknown	4	27	7	29	11	28
MYCN status						
Not amplified	7	47	11	46	18	46
Amplified	4	27	2	8	6	15
Unknown	4	27	11	46	15	38
Ploidy						
Hyperdiploid	6	40	10	42	16	41
Diploid	4	27	3	12	7	18
Unknown	5	33	11	46	16	41
Histology						
Favorable	0	0	0	0	0	0
Unfavorable	9	60	11	46	20	51
Unknown	6	40	13	54	19	49
No. of courses administered						
Total	35		76		110	
Median	2		2.5		2	
Range	1-6		1-6		1-6	

Abbreviation: INSS, International Neuroblastoma Staging System.
*Disease measurable by standard radiographic criteria.
†Disease evaluable only by iodine-123 metaiodobenzylguanidine and/or bone marrow histology.

Table 2. Response Summary

Stratum	No. of Evaluable Patients	No. of Responders	Level of Response				
			CR	VGPR	PR	SD	PD
1 (n = 15)	13	0	0	0	0	3	10
2 (n = 24)	23	5	5	0	0	4	14

Abbreviations: CR, complete response; VGPR, very good partial response; PR, partial response; SD, stable disease; PD, progressive disease.

patients responded of the first 20 evaluable in a given stratum, the regimen was considered effective.

A two-stage rule was used to monitor for an excessive number of unacceptable DLTs, where unacceptable was defined as a requirement for pressor and/or ventilator support due to acute vascular leak syndrome. Secondary analyses of EFS and OS were performed as intent to treat. For EFS, time to event was from enrollment until first occurrence of relapse, progression, death, or secondary malignancy or until last contact if no event was observed. For OS, the event was death. Survival estimates (Kaplan-Meier) were calculated[40] and reported with SEs.[41]

Estimates of the mean value of biologic correlates are presented ± the SE. A paired *t* test was used to test the change from baseline to a subsequent time point. A two-sample *t* test was used to compare the level of a particular biologic correlate for responders versus nonresponders. A nonparametric Spearman's rank correlation analysis was performed to test for association between hu14.18-IL2 levels and anti-hu14.18-IL2 antibody response (both the bridging and the binding assays). All analyses were performed using SAS software version 9.2 (SAS Institute, Cary, NC). *P* values less than .05 were considered statistically significant.

RESULTS

Patient Characteristics

A total of 39 patients (all eligible) were enrolled, 15 in stratum 1 and 24 in stratum 2 (Table 1). The 15 patients in stratum 1 received a total of 35 treatment courses (median, two courses), and the 24 patients in stratum 2 received a total of 76 courses (median, 2.5 courses).

Response and Outcome

Two patients in stratum 1 were not evaluable for response. One received no treatment due to parental choice, and the other received only one dose of drug secondary to vascular leak and hypotension. Of the 13 evaluable patients in stratum 1, there were no responders: three had SD and 10 had PD. One patient in stratum 2 was taken off study secondary to anaphylaxis during cycle 1 and was not evaluable for response, leaving 23 evaluable stratum 2 patients. In the first 20 evaluable stratum 2 patients, there were five responders, all with CR (Table 2). The statistical criterion for activity required at least four responders in stratum 2, and this boundary was exceeded. Of the 23 evaluable stratum 2 patients, five patients had a CR, four patients had SD, and 14 had PD, for an overall response and CR rate of 21.7% (95% CI, 5% to 37%).

Three of the patients with CR (Table 3) enrolled with disease in the BM only. One patient had a single MIBG-avid lesion in the right tibia, and the final responder had BM disease as well as multiple MIBG-avid sites. This was the first relapse for four of the five patients who had previously been in a complete remission after myeloablative chemotherapy and autologous stem-cell transplantation (ASCT). Patient 29 had primary refractory neuroblastoma and enrolled with persistent disease 2 months after treatment with ^{131}I-MIBG and myeloablative therapy with autologous stem-cell rescue. Four of these five patients received six cycles of therapy, and one (patient 10) stopped therapy after four cycles due to DLT. Two of the responders received isotretinoin after the completion of protocol-determined therapy. Four of the patients achieved CR after two cycles of hu14.18-IL2 treatment. Patient 29 had a negative MIBG scan and negative BM morphology after two cycles of treatment but remained positive by immunocytology. Both the BM morphology and immunocytology were clear after four treatment cycles. All five patients had a prolonged CR, and patient 29 remains in CR at 35+ months (additional clinical details for these patients are provided in Appendix Table A1, online only).

In addition to the five CRs, two additional patients in stratum 2 who were scored as having SD for protocol-defined agent activity showed suggestion of improvement and are presented here descriptively (patients 3 and 21 in Appendix Table A1). One patient went on study with multiple MIBG-avid sites and biopsy-proven bone and marrow disease after ASCT. This patient showed clearing of marrow disease and had a decrease in MIBG avidity that was close to, but did not meet, the definition of PR by central review. The other patient went on study with MIBG-avid disease and BM biopsies showing 10% to 15% replacement with neuroblastoma. After four courses of treatment, despite a CR by MIBG scintigraphy, the overall response was SD because of substantial improvement, but incomplete clearing in the BM.

The overall (n = 39) 1-year EFS and OS were 26% ± 10% and 63% ± 11%, respectively, with the curves going much lower after 1 year (Fig 1A). For stratum 1 (n = 15) and stratum 2 (n = 24), both the

Table 3. Response Details

Patient	Disease at Study Entry	Courses	Dose Reduction Required	Response	Time to Event* (months)
2	Bone marrow	6	No	CR	13†
10	Bone marrow	4	Yes	CR	9
22	MIBG (1 site)	6	Yes	CR	20†
27	Bone marrow	6	No	CR	30
29	Bone marrow, MIBG (multiple sites)	6	No	CR	No event‡

Abbreviations: CR, complete response; MIBG, iodine-123 metaiodobenzylguanidine.
*Time to progression from start of therapy.
†Patient received *cis*-retinoic acid after the completion of hu14.18-IL2.
‡Patient in remission for 35 months at last follow-up.

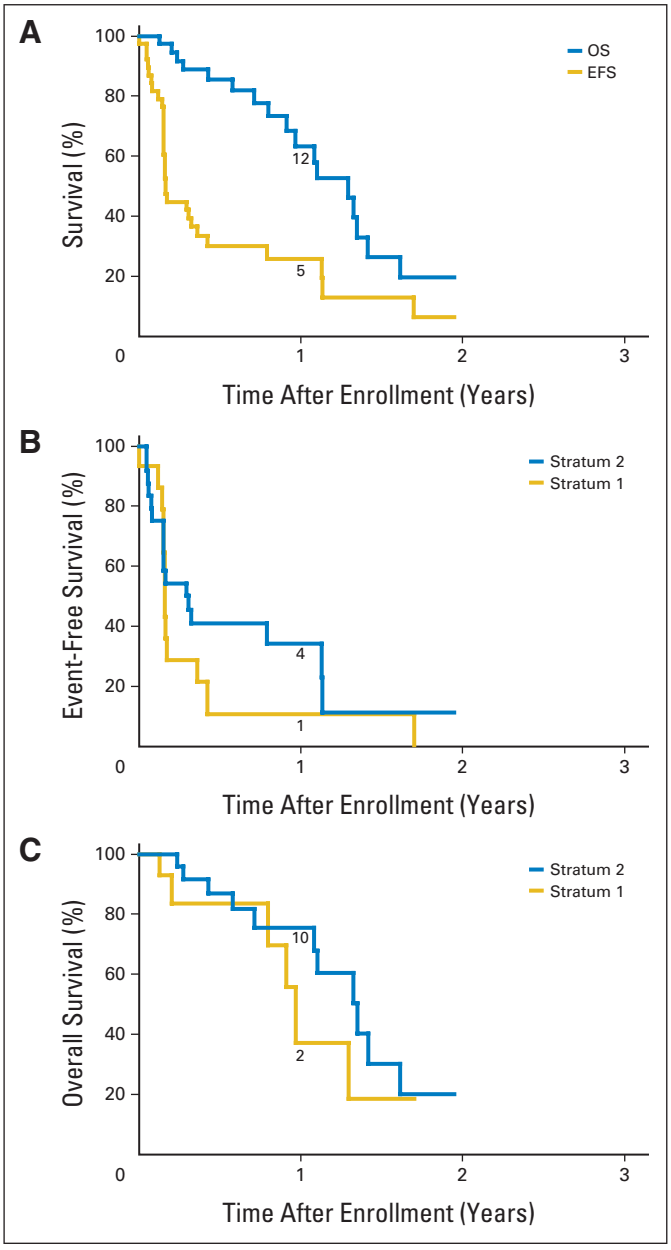

Fig 1. (A) Event-free survival (EFS) and overall survival (OS) for all patients; (B) EFS for stratum 1 and stratum 2; (C) OS for stratum 1 and stratum 2. The numbers alongside each curve, at the 1-year time point, indicate the number of patients corresponding to that curve at the 1-year time point.

EFS (Fig 1B) and OS (Fig 1C) curves trend to similar low values after 1 year.

Toxicity

Of the 38 patients evaluable for toxicity, eight received only one course of therapy: six due to PD and two due to DLT. The grade 3 and 4 toxicities observed over all treatment courses are listed in Table 4. Most toxicities were self-limited and resolved within a few days of the last dose of hu14.18-IL2 for that treatment course.

Two patients had unacceptable DLTs. One developed grade 3 hypotension after the first dose of hu14.18-IL2 in course 1 and required treatment with dopamine for 24 hours. The other developed capillary leak and hypoxia that required pressors and ventilator support for 2 weeks. This toxicity developed after the final dose of hu14.18-IL2 during course 2. In retrospect, this patient had two prior episodes requiring ventilator support because of capillary leak after ASCT 1 year prior. After this event, the protocol was amended to exclude patients with a prior history of ventilator support related to lung injury. All DLTs are listed in Table 5.

Table 4. Grade 3 and 4 Toxicities for All Courses of Therapy

Toxicity	No. of Patients (n = 38)*	Incidence of Toxicity (%)
Acute vascular leak syndrome	12	31.6
Allergic reaction/hypersensitivity	4	10.5
ALT elevation	8	21.1
AST elevation	9	23.7
Bilirubin	8	21.1
Fever (without neutropenia)	15	39.5
Hemoglobin	9	23.7
Hypokalemia	4	10.5
Hyponatremia	2	5.3
Hypotension	6	15.8
Infection (catheter-related) with ANC > 1,000/μL	5	13.2
Leukocytes	9	23.7
Lymphocytes	15	39.5
Neutrophils	13	34.2
Pain (head/headache)	4	10.5
Pain (other)	12	31.6
Platelets	16	42.1
Pleural effusion (nonmalignant)	2	5.3
Pneumonitis/pulmonary infiltrates	2	5.3
Rash	2	5.3
Urticaria	2	5.3

Abbreviation: ANC, absolute neutrophil count.
*Number of patients reporting at least one grade 3 or 4 toxicity over all courses. Treatment was not initiated in one patient.

Correlative Studies

Stratum 1 and stratum 2 patients were combined for these correlative analyses.

Hu14.18-IL2 levels. The mean change in the serum hu14.18-IL2 level from baseline (course 1, day 1, before first dose) to (1) the day 1 peak value was 2.4 $\pm$ 0.9 μg/mL (n = 36) and (2) the day 3 peak value was 2.1 $\pm$ 0.8 μg/mL (n = 31). During course 1, the change from baseline to day 3 was less than the change from baseline to day 1 ($P < .001$); this was true for all courses (courses 1 through 6). Within the 36 patients evaluable for response, for each time point (day 1 peak, day 3 peak) and course (1 through 6), the hu14.18-IL2 peak levels for responders (n = 5) were similar to those of nonresponders ($P > .15$ at each time).

Absolute lymphocyte count. As noted previously,[34] subjects showed a significant ($P < .001$) decrease in their absolute lymphocyte count (ALC) with hu14.18-IL2 treatment (course 1, baseline to day 3 decrease of 830 $\pm$ 940 cells/μL [n = 29]; baseline to day 4 decrease of 710 $\pm$ 770 cells/μL [n = 25]). Although this drop in ALC is scored as hematologic toxicity, it actually represents immune activation and margination of lymphocytes, a known effect of IL-2.[42] This transient lymphopenia (Appendix Fig A1, online only) is followed by lymphocytosis consistent with immune activation (course 1, baseline to day 8 increase ($P < .001$) of 2,360 $\pm$ 2,160 cells/μL [n = 26]). A similar

Table 5. Dose-Limiting Toxicity

Patient	Course	Toxicity	Result
3	2	Grade 3 hypoxia, pneumonitis/pulmonary infiltrates	Tolerated courses 3-6 at 50% dosing
4	2	Grade 4 acute vascular leak	Therapy discontinued due to toxicity
10	3	Grade 3 acute vascular leak	Course 4 at 50% dosing, discontinued day 2
13	1	Grade 3 acute vascular leak and hypotension	Therapy discontinued due to toxicity
14	2	Grade 3 hyperbilirubinemia	Course 3 50% dosing, course 4 25% dosing
18	1	Grade 3 transaminitis	Tolerated course 2 at full dose*
19	3	Grade 3 transaminitis	Tolerated course 4 at 50% dosing
21	4	Grade 3 transaminitis	Repeat toxicity course 5 at 50% dosing
22	4	Grade 3 hyperbilirubinemia	Tolerated course 5 at 50% dosing and course 6 at 75% dosing
24	1	Grade 3 transaminitis	Tolerated courses 2-4 at 50% dosing
26	1	Grade 3 hyperbilirubinemia	Tolerated course 2 at 50% dosing
31	2	Grade 3 hypotension	Off study end of course due to PD
32	1	Grade 4 allergic reaction	Therapy discontinued due to toxicity
34	2	Grade 3 hypotension	Tolerated course 3 at 50% dosing and course 4 at 75% dosing
37	1	Grade 3 transaminitis	Tolerated course 2 at 50% dosing
38	2	Grade 3 acute vascular leak	Tolerated courses 3-6 at 50% dosing

Abbreviation: PD, progressive disease.
*Dosing in violation of the protocol.

pattern of somewhat smaller ALC decreases from baseline to days 3 and 4 was seen in subsequent courses; the decreases in courses 5 and 6 were not significant.

sIL2R levels. As noted previously,[34] there was a significant increase in sIL2R levels at all courses from baseline to days 4 and 8 ($P < .0001$ for courses 1 through 3; $P < .01$ for courses 4 through 6). sIL2R values in courses 2, 3, 5, and 6 were higher than on corresponding days in course 1. Within the 36 patients evaluable for response, 31 reported an sIL2R level on day 4 of course 1: the five responders had a mean sIL2R of 17,006 $\pm$ 6,277 pg/mL versus 11,104 $\pm$ 4,372 pg/mL for the 26 not responding ($P = .015$). In a comparison of sIL2R levels for the patients with a DLT versus those without a DLT, there was no association.

Anti-hu14.18-IL2 antibody response. Of 36 evaluable patients, 13 patients developed an anti-idiotypic antibody against hu14.18-IL2 based on the bridging assay, and 16 developed an anti-idiotypic antibody based on the binding inhibition assay.[38,39] However, there was no apparent effect of this anti-idiotypic antibody response on the in vivo level of hu14.18-IL2. Specifically, there was no significant association of the level of anti-idiotypic antibody developed after course 1 (or after course 2) with any detectible decrease in peak hu14.18-IL2 level seen on day 1 of course 2 versus the level seen on day 1 of course 1. This is in contrast to the decrease in hu14.18-IL2 levels from course 1, day 1, to course 2, day 1, for those patients with a strong anti-idiotypic antibody response in our past phase I trials (where most patients received lower doses).[39] Furthermore, there was no association of anti-idiotypic antibody response (by either of these assays) with antitumor effect for the five CRs.

All of the correlative analyses described above comparing the five patients in CR with the others were repeated, comparing the seven "improved" patients (ie, the five patients with CRs plus the two patients in stratum 2 who were scored as having SD, but showed clinical improvement in BM and or MIBG [patients 3 and 21 in Appendix Table A1]) versus the other patients. For this comparison, no statistically significant associations were found between hu14.18-IL2 levels, sIL2R levels, or anti-idiotypic antibody response with antitumor activity. Furthermore, no significant associations were found between response and factors at diagnosis (age, stage, *MYCN*, ploidy, or histologic grade; Appendix Table A1).

DISCUSSION

This study demonstrates antitumor activity of hu14.18-IL2 in patients with relapsed/refractory neuroblastoma with stratum 2 disease. Five (of 23 evaluable) stratum 2 patients had a durable CR to therapy, and two additional patients showed evidence of improvement. Although this study did not collect data specifically quantifying disease burden at enrollment, there is the suggestion from their clinical descriptions that the five responders began treatment with relatively small but clearly evaluable tumor burdens: limited MIBG-avid lesions (rather than diffuse skeletal MIBG avidity) and partial contamination of marrow with tumor cells (rather than marrow replacement). Even so, all responders had a poor clinical prognosis after being refractory to or relapsing after frontline therapy. In contrast, none of the 15 patients entered into stratum 1 showed evidence of antitumor activity. This trial was not designed or powered to test for a difference in the response rate between stratum 1 and 2; however, five CRs of 23 evaluable patients in stratum 2 compared with 0 of 13 patients in stratum 1 has a *P* value of .089. If one includes in this analysis the two additional stratum 2 patients with SD but descriptive improvement (patients 3 and 21 in Appendix Table A1), the difference is significant between the strata ($P = .029$). These results are consistent with preclinical data showing that the efficacy of hu14.18-IL2 is best seen when used in the minimal residual disease setting.[14]

The clinical toxicities seen in this study were consistent with those previously reported for hu14.18-IL2[33,34] and for anti-GD2 mAb plus IL-2.[19-21,25] Most toxicities resolved within days; only three patients had their therapy discontinued because of toxicity.

Evidence for immune activation was seen as changes in sIL2R levels and lymphocytosis. Neither of these were correlated with antitumor response or with toxicity. Although there was a significant increase in sIL2R levels in the five responders compared with the others, this correlation was not seen when the two "improved" patients

were included in the analysis. Anti-idiotypic antibody was detected in 13 and 16 of 36 patients using two different assays. This anti-idiotypic antibody was not correlated with antitumor activity, in contrast to clinical response correlations with human antimouse antibody detection reported in other studies.[43,44] This may be due in part to low statistical power in this study. Furthermore, the anti-hu14.18-IL2 responses we detected did not seem to have functional significance in that they were not associated with a subsequent decrease in hu14.18-IL2 levels. This suggests that the anti-idiotypic antibodies detected were not sufficiently strong to impact the function of the circulating hu14.18-IL2.

The results of this study support further development of hu14.18-IL2 in patients with recurrent or refractory neuroblastoma with disease evaluable only by ^{123}I-MIBG scintigraphy and/or BM histology. A successor study is being planned to confirm efficacy in stratum 2 patients and quantify the disease burden in patients before and after treatment to better define which patients are most likely to respond to hu14.18-IL2 (see Appendix, online only).

Finally, given the efficacy recently demonstrated for the regimen of ch14.18 mAb plus IL-2 plus GM-CSF for children with high-risk neuroblastoma who have achieved response (CR, very good PR, or PR) to their initial induction and consolidation treatment[27] and the superiority of ch14.18-IL2 over ch14.18 plus IL-2 as separate molecules in preclinical studies (Buhtoiarov et al, manuscript submitted for publication; Gubbels et al, manuscript submitted for publication),[28-30] we hypothesize that hu14.18-IL2 may be more effective than ch14.18 plus IL-2 in this same clinical setting. Thus the COG is planning to randomly compare a regimen of hu14.18-IL2 plus GM-CSF plus isotretinoin versus the now "standard" regimen of ch14.18 plus GM-CSF plus IL-2 plus isotretinoin in a phase III study for newly diagnosed patients with high-risk neuroblastoma who have achieved response to their front-line therapy.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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Glossary Terms

Monoclonal antibody: An antibody that is secreted from a single clone of an antibody-forming cell. Large quantities of monoclonal antibodies are produced from hybridomas, which are produced by fusing single antibody-forming cells to tumor cells. The process is initiated when a mouse is immunized initially against a particular antigen, stimulating the production of antibodies targeted to different epitopes of the antigen. Antibody-forming cells are subsequently isolated from the spleen. By fusing each antibody-forming cell to tumor cells, hybridomas can be generated each with a different specificity and targeted against a different epitope of the antigen

ADCC (antibody-dependent cell-mediated cytotoxicity): a mechanism of cell-mediated immunity whereby an effector cell of the immune system actively lyses a target cell that has been bound by specific antibodies.

Hu14.18-IL2: an immunocytokine, which is a fusion protein, comprised of one molecule of humanized anti-GD2 monoclonal antibody, with an intact molecule of human interleukin-2 on the carboxy terminus of each immunoglobulin G heavy chain.

MIBG Scintigraphy: a nuclear medicine scan using iodine-123 metaiodobenzylguanidine (MIBG) scintigraphy to identify neuroblastoma or pheochromocytoma lesions.