Gallium-67 and Technetium-99m-Methylene Diphosphonate Skeletal Scintigraphy in Determining Prognosis for Children with Stage IV Neuroblastoma

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Thirty-five children (aged 0–9 yr) who had presented with Stage IV neuroblastoma were studied to see if avidity for ^{67}Ga or ^{99m}Tc-methylene diphosphonate (MDP) uptake in both primary and secondary sites at diagnosis conferred any prognostic significance. Twenty-three percent of the patients were disease free and off treatment at the time of study. Crude survival did not differ between groups. Duration of survival and the likelihood of completing treatment were related to the scintigraphic appearance at the time of diagnosis, after adjustment for potential confounding effects, using Cox's proportional hazards regression and multiple logistic regression. After adjustment for confounding influences, neither ^{67}Ga avidity nor uptake of ^{99m}Tc-MDP was associated with a significantly worse prognosis, both in terms of adjusted survival and likelihood of completing treatment. Patients with ^{67}Ga-avid scans at diagnosis did not demonstrate significantly worse survival (HR 1.47, 95% CI 0.43–5.11) than those without ^{67}Ga avidity. They were somewhat less likely to complete treatment (OR 0.23, 95% CI 0.03–1.63), but this did not reach statistical significance. Similarly, although patients with ^{99m}Tc-MDP positive scans demonstrated somewhat worse survival (HR 2.47, 95% CI 0.45–13.54), this result did not reach statistical significance, nor were they less likely to complete treatment (OR 0.69, 95% CI 0.07–6.67) than those with ^{99m}Tc-MDP negative scans. Uptake of ^{99m}Tc-MDP into extraosseous sites was also not associated with worse survival (HR 1.45, 95% CI 0.58–3.62) nor with decreased likelihood of completing treatment (OR 0.78, 95% CI 0.12–5.09). Other than indicating disease stage, these results do not support the hypothesis that the scintigraphic appearance at diagnosis confers prognostic information in children with advanced neuroblastoma.

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Next to brain tumors, neuroblastoma is the most common solid malignancy of childhood, accounting for approximately 8% of all childhood cancers (*1,2*). It is also the most common malignancy in infancy (*2*).

A staging system for neuroblastoma was proposed 20 yr ago for the Children's Cancer Study Group (CCSG) by Evans, D'Angio and Randolph (*3*) and is the one in common use in Australia today, although a new, internationally standardized staging system has recently been proposed (*4*). More than half of children with neuroblastoma present with advanced (Stage IV) disease (*1*).

Survival in children presenting with neuroblastoma is related to a number of factors, of which the most informative appear to be age and stage at diagnosis. Younger patients, particularly those less than 1 yr old, and those with localized disease have a better prognosis (*1,2*). Other parameters reported to have prognostic significance include site of primary, tumor histology, catecholamine profile, serum ferritin, serum neuron-specific enolase, chromosome pattern and amplification of the N-myc oncogene (*5–10*).

Results of treatment of advanced neuroblastoma with surgery, chemotherapy and radiotherapy have been disappointing. The overall 2-yr survival rate is reported to be between 7% and 23% and has not increased dramatically over the past 30 yr (*11–13*). In an effort to improve this situation, new treatment modalities have been developed. In particular, bone marrow transplantation in first remission, treatment with radioiodinated meta-iodobenzylguanidine (MIBG) and more intensive induction chemotherapy have shown promise (*14–18*). It is evident that there is some heterogeneity among patients with disseminated neuroblastoma and that some patients may do better with conventional therapies than others (*1,2,16*). Accurate means of assessing prognosis have become more important as treatment options have broadened.

Extra-osseous uptake of ^{99m}Tc-methylene diphosphonate (MDP) is said to be pathognomonic of neural crest tumors in childhood (*19*), but only about two-thirds of

neuroblastomas will exhibit this phenomenon (*20–22*). Skeletal metastases are evident with ^{99m}Tc-MDP scanning in 40%–60% of childhood neuroblastoma overall (*19,20*) and this is associated with a worse prognosis (*23*), although this may merely reflect more advanced disease at diagnosis.

Gallium-67-citrate scanning may also be useful in the initial evaluation of patients with neuroblastoma but is inferior to ^{99m}Tc-MDP for the detection of skeletal metastases (*22,24,25*). Some studies have shown that the degree of uptake of ^{67}Ga by the tumor cells has prognostic significance (*24,26,27*), but the number of patients in each study has been small.

We analyzed the records of patients treated for Stage IV neuroblastoma in the oncology unit at this institution in order to see whether ^{67}Ga avidity or ^{99m}Tc-MDP uptake into primary tumors and metastases had any prognostic significance in our patient population.

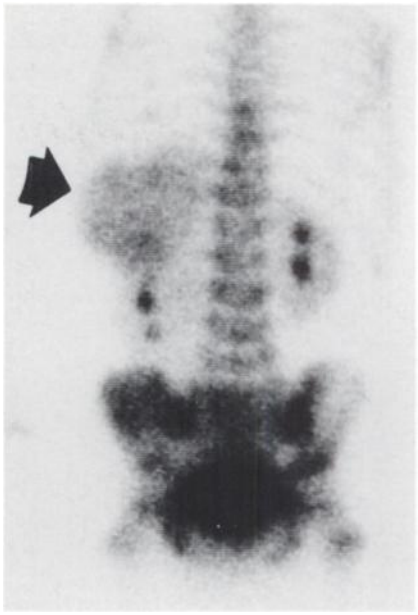

FIGURE 1. Technetium-99m-MDP bone scan. (Posterior view) There is avid uptake of tracer into the primary neuroblastoma in the left suprarenal area (arrow). Note metastatic disease in the vertebrae, pelvis and proximal left femur.

MATERIALS AND METHODS

All children admitted to the oncology unit between February 1975 and December 1988 with a diagnosis of Stage IV neuroblastoma according to the CCSG staging system (*3*) were included in the study. Patients admitted after this period were excluded because the first line treatment of this disease changed at the beginning of 1989 to one of intensive chemotherapy and bone marrow transplantation in first remission. The diagnosis was determined in most cases by histological examination of biopsy specimens from the primary tumor or metastases. Some patients had been diagnosed without biopsy where there was elevated urinary excretion of catecholamines typical of neuroblastoma in association with radiographic or radionuclide evidence of widespread metastatic disease. Patients were classified as Stage IV when there was evidence of metastases in bone (on bone scan) or in bone marrow or solid organs (on biopsy). Patients with Stage IV-S disease were excluded.

All patients were treated with combination chemotherapy (vincristine, cyclophosphamide, dacarbazine and doxorubicin) in monthly cycles for 2 yr. Excision of the primary tumor, where feasible, was attempted in some patients who had achieved a good partial or complete remission.

Some patients received radiotherapy either to the tumor bed postoperatively or to troublesome metastases. Three patients received HLA-compatible allogeneic bone marrow transplants after failure of conventional treatment, although this was not first line treatment at this time.

Total body bone scans were performed by injection of ^{99m}Tc-MDP in doses calculated by weight according to an adult reference dose of 740 MBq. Dynamic and blood-pool scans were performed initially and whole-body bone scans performed at 2 hr on a LFOV gamma camera using a high-resolution, low-energy, parallel-hole collimator (Fig. 1).

Gallium scans were performed by injection of ^{67}Ga-citrate in doses calculated by weight according to an adult reference dose of 185 MBq. Whole-body scans were performed at 48 hr postinjection, and again at 72 hr if necessary, using triple pulse-height analysis and a medium-energy, parallel-hole collimator (Fig. 2).

The results were determined by a review of the patients' notes and radionuclide scans. Primary tumors and metastases were said to exhibit either ^{99m}Tc-MDP uptake and/or ^{67}Ga avidity if the scans showed a nonphysiological accumulation of tracer which could be related to the presence of tumor either in the primary or in sites of metastatic disease.

The data were analyzed with the statistical package EGRET (Statistics and Epidemiological Research Corporation, Seattle, WA). The method of Kaplan and Meier was used to determine crude survival curves for each category of patient and any differences were assessed using the log rank test. Cox's proportional hazards regression was used to assess survival after adjustment for the confounding variables, which were age at diagnosis, sex, site of primary and treatments employed, and any significant interactions (alpha = 0.05). Logistic regression was used to determine the likelihood of completing treatment after adjustment for the same confounding variables. Fisher's exact test was used to determine differences in proportions and differences in means were assessed using Student's t-test. All tests were two-tailed.

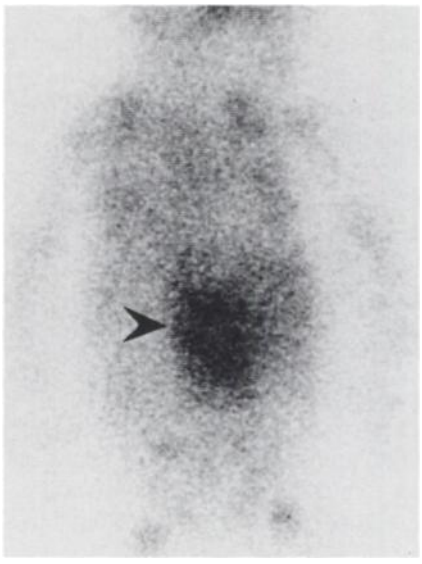

FIGURE 2. Gallium-67 study. (Anterior view) There is avid uptake of ^{67}Ga in the large abdominal neuroblastoma (arrow).

TABLE 1
Characteristics and Management of 35 Children with Stage IV Neuroblastoma

	Survivors (n = 8)	Nonsurvivors (n = 27)	Fisher's exact test
Sex			
Male	3	16	
			p = 0.50
Female	5	11	
Age			
<1 yr	3	3	
			p = 0.23
>1 yr	5	24	
<2 yr	8	7	
			p = 0.0005
>2 yr	0	20	
Site*			
Abdominal	7	24	
			p = 1.0
Extra-abdominal	1	2	
Treatment			
Surgery	6	4	
			p = 0.0056
No surgery	2	23	
Radiotherapy	4	3	
			p = 0.067
No radiotherapy	4	24	
Transplant	2	1	
			p = 0.25
No transplant	6	26	

*The site of primary tumor could not be determined in one patient.

RESULTS

Thirty-five patients were included in the study. Two patients who were still undergoing treatment were excluded. The age range of patients was 1 mo to 9 yr. The median survival of all patients was 14 mo (95% confidence interval 10–20 mo). Of the 35 patients included, 8 (23%) were still alive and disease-free at the time of study (survivors) with a mean duration of survival of 87.5 mo (range 36–140 mo).

Patient characteristics are presented in Table 1. Survivors had a significantly lower mean age at diagnosis (13 mo) than nonsurvivors (40 mo) ($p < 0.001$), and significantly more survivors presented less than 2 yr old compared to nonsurvivors ($p < 0.001$). No patient presenting at greater than 2 yr old survived.

Significantly more survivors were treated with surgery than were nonsurvivors ($p < 0.001$). Similarly, more survivors received radiotherapy than did nonsurvivors but this difference was not significant ($p = 0.067$). Radiotherapy was given as an adjunct to surgery in all but three cases. One of these was a patient with thoracic neuroblastoma and the other two were patients with terminal disease and who had withdrawn from active treatment. Two survivors (25%) received bone marrow transplants compared to one nonsurvivor (4%), but this difference was not significant ($p = 0.25$). The site of primary was identified in all survivors but could not be established in one nonsurvivor. Twelve patients completed treatment. Of these, four (33%) subsequently relapsed and died. All did so quickly, with a mean time from completion of therapy to relapse of 4.5 mo (range 1–8 mo).

TABLE 2
Median Survival Times (Crude) in Relation to Radionuclide Scans

	No.	Median survival (mo)	Log rank test
^{99m}Tc-MDP negative	3	32	
			p = 0.98
^{99m}Tc-MDP positive	32	14	
^{99m}Tc-MDP uptake into bone only	20	14	
			p = 0.98
^{99m}Tc-MDP uptake into bone and soft tissues	12	14	
^{67}Ga nonavid	7	27	
			p = 0.42
^{67}Ga avid	22	11	

Radionuclide Bone Scans

All 35 patients were studied with ^{99m}Tc-MDP scans at diagnosis. Only three patients (9%) had bone metastases that failed to take up ^{99m}Tc-MDP but which were evident using conventional radiography. The crude survival and adjusted hazard ratios for children with and without uptake of ^{99m}Tc-MDP are presented in Tables 2 and 3, respectively, whereas the likelihood of completing treatment for both groups after adjustment for possible confounding is

TABLE 3
Adjusted Survival in Relation to Radionuclide Scans

	Hazard ratio	95% confidence interval
^{99m}Tc-MDP negative	1.00	—
^{99m}Tc-MDP positive	2.47	(0.45, 13.54)
^{99m}Tc-MDP uptake into bone only	1.00	—
^{99m}Tc-MDP uptake into bone and soft tissues	1.45	(0.58, 3.62)
^{67}Ga nonavid	1.00	—
^{67}Ga avid	1.47	(0.43, 5.11)

TABLE 4
Probability of Completing Treatment in Relation to Radionuclide Scans

	Odds ratio	95% confidence interval
^{99m}Tc-MDP negative	1.00	—
^{99m}Tc-MDP positive	0.69	(0.07, 6.67)
^{99m}Tc-MDP uptake into bone only	1.00	—
^{99m}Tc-MDP uptake into bone and soft tissues	0.78	(0.12, 5.09)
^{67}Ga nonavid	1.00	—
^{67}Ga avid	0.23	(0.03, 1.63)

presented in Table 4. There were no statistically significant differences in any of these parameters between those with ^{99m}Tc-MDP positive scans and those with negative scans, although the small number of children with ^{99m}Tc-MDP negative scans at diagnosis did exhibit longer survival.

Fourteen patients (40%) exhibited uptake of ^{99m}Tc-MDP into the primary tumor or nonbony metastases (extra-osseous uptake). There was no difference in median survival between those with osseous uptake only and those with extra-osseous uptake (Table 2). Similarly, after adjustment for the confounding variables, no significant differences in hazard ratio (Table 3) or odds ratio for completing treatment (Table 4) were found between those children with osseous uptake only and those with extra-osseous uptake.

Gallium-67-Citrate Scans

Twenty-nine patients were studied with ^{67}Ga scans at diagnosis. Median survival in patients with gallium-avid tumors was shorter than in those whose tumors were not gallium-avid (11 versus 27 mo, respectively), although this difference was not significant (log rank test $p = 0.42$). No difference in overall survival was seen between the two groups after adjustment for confounding variables (Table 3). Patients with ^{67}Ga-avid tumors were somewhat less likely to complete treatment than those with nonavid tumors, although, once again, this difference did not reach statistical significance (Table 4).

DISCUSSION

The overall survival of our patients is similar to that presented in two recent series of patients receiving conventional treatment modalities (*11,28*). All of our survivors were more than 24 mo from completion of treatment. Two of the survivors received successful bone marrow transplants in remission, and this may have improved the survival figures beyond those expected with other, more conventional therapies but the number in our series is too small to allow a valid analysis of this effect.

As expected, the survivors were much younger on average than the nonsurvivors. Some authors have also reported improved survival in the small number of older patients (>8 yr old) presenting with Stage IV neuroblastoma (*29,30*) but there was only one patient in our series who was more than 8 yr old at diagnosis, a 9-yr-old male who died 15 mo following diagnosis. None of our patients presenting at more than 2 yr old survived and this concurs with another large study which showed no survivors in those presenting greater than 12 mo with Stage IV disease (*10*).

The number of patients receiving surgery and/or radiotherapy was greater among the survivors. Our policy was to offer these treatment modalities only to patients who achieved at least a good partial remission with combination chemotherapy, and it is not surprising that there were more of these among the survivors.

Technetium-99m-MDP uptake was observed in bony metastases in 91% of our patients. This conforms to previously published data (*19,20,22,31*). The primary tumor was visible in 40% of scans. This is less than the generally quoted figure of two-thirds (*20–22*) but is in agreement with some studies (*19*). Uptake of ^{99m}Tc-MDP was not useful for predicting long-term survival, except as a means of assessing disease stage. There were no survivors from the group with ^{99m}Tc-MDP negative studies, although this group was somewhat more likely to complete treatment and demonstrated better short-term survival (lower hazard ratio). None of these results, however, were statistically significant. Patients with extra-osseous uptake of ^{99m}Tc-MDP fared no better or worse than those with uptake into bony metastases only. This result concurs with a previously published study (*32*).

The percentage of patients in our series with ^{67}Ga-avid tumors (*76%*) is higher than that reported in some other series (*27,33,34*), but these series involved small numbers of patients and were not controlled for stage of disease. All of our patients had Stage IV disease, which may be associated with greater ^{67}Ga avidity, and improved scanning techniques in recent years have resulted in better image quality (*35*). Another, more recently published series of 14 patients, also uncontrolled for stage of disease, indicated ^{67}Ga-avid tumors in 79% (*22*).

Gallium-67 is postulated to enter tumor cells by acting as a ferric ion analogue. It is incorporated into the transferrin molecule, which then binds to receptors on the cell membrane (*36,37*). Intracellular dissociation of ^{67}Ga from the transferrin molecule is facilitated by an acid pH (*38*); gallium avidity may be higher in those tumors with low intracellular pH, although this has not been studied as a prognostic factor. The factors influencing iron metabolism in neuroblastoma are not clear. The serum ferritin level is highest in patients with more advanced (Stage III and IV) neuroblastoma, but it is also a prognostic indicator in its own right, independent of disease stage (*5,9*). Patients with ^{67}Ga-nonavid tumors were somewhat more likely to complete treatment and had a longer duration of survival, but the overall survival was not related to ^{67}Ga avidity in our

patients. It is possible that there is more than one disturbance of iron metabolism in neuroblastoma cells.

The number of patients in this study, although larger than in any previously published work that has examined this issue, was small and it is doubtful if any one treatment center would have seen sufficient patients to produce analyses with sufficient statistical power to conclusively demonstrate an effect. These results suggest that neither avidity for ^{67}Ga nor extra-osseous uptake of ^{99m}Tc-MDP is useful as a means of determining prognosis for children with Stage IV neuroblastoma, but further study using large numbers of patients from multiple centers seems desirable. Technetium-99m-MDP scanning remains useful in all patients as a sensitive means of staging disease.

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