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ORIGINAL ARTICLE

Resection of primary tumor in stage 4S neuroblastoma: a second study by the Italian Neuroblastoma Group

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Abstract

Purpose To clarify the role of primary tumor resection in stage 4S neuroblastoma.

Methods We investigated a cohort of 172 infants diagnosed with stage 4S neuroblastoma between 1994 and 2013. Of 160 evaluable patients, 62 underwent upfront resection of the primary tumor and 98 did not.

Results Five-year progression-free and overall survival were significantly better in those who had undergone upfront surgery (83.6% vs 64.2% and 96.8% vs 85.7%, respectively). One post-operative death and four non-fatal complications occurred in the resection group. Three patients who had not undergone resection died of chemotherapy-related toxicity. Thirteen patients underwent late surgery to remove a residual tumor, without complications: all but one alive. Outcomes were better in patients diagnosed from 2000 onwards.

Conclusion Infants diagnosed with stage 4S neuroblastoma who underwent upfront tumor resection had a better outcome. However, this result cannot be definitely attributed to surgery, since these patients were selected on the basis of their favorable presenting features. Although the question of whether to operate or not at disease onset is still unsolved, this study confirms the importance of obtaining enough adequate tumor tissue to enable histological and biological studies to properly address treatment, to achieve the best possible outcome.

Keywords Neuroblastoma · Stage 4S · Surgery · Primary tumor resection

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Introduction

The term stage 4S neuroblastoma refers to infants up to 1 year of age who are diagnosed with a localized primary tumor associated with remote disease that is confined to liver, skin, and/or bone marrow (< 10% infiltration) [1]. Its natural history is characterized by a period of tumor progression (lasting from a few days to some months) that may lead to death regardless of therapy, or be followed by therapy-induced or spontaneous regression [2], the mechanism of which is not fully understood [3]. The probability of cure is fairly high and has increased from 60% in the1980s [4–7] to the present 90% [8–12].

The therapeutic approach to stage 4S neuroblastoma is not well defined, in particular for what concerns the role of resection of the primary tumor. Two studies have focused on this issue: back in 1992, Martinez et al. analyzed 37 such infants and concluded that resection was associated with a better outcome [13]. A few years later, however, Guglielmi et al. were unable to confirm this favorable effect in a study of 94 Italian patients [14]. Other authors have expressed divergent opinions on the issue. For example, Stokes et al. [5], Blatt et al. [7], and Katzenstein et al. [8] stated that resection of the primary did not correlate with survival, while Berthold et al. [15] maintained that it could improve outcome, and Evans et al. [4] and Nickerson et al. [9] advocated primary resection to prevent local recurrence. Finally, a recent Children's Oncology Group (COG) study suggested that primary resection could be avoided in symptomatic patients requiring emergency chemotherapy [16]. In an attempt to provide new useful information on the question of the advantage of primary tumor resection in infants diagnosed with stage 4S neuroblastoma, we retrospectively analyzed the records of a large cohort of such infants diagnosed in Italy in the 20-year period following the previous Italian report on this issue [14].

Methods

Between 1994 and 2013, a total of 2310 subjects aged 0–18 years with previously untreated neuroblastoma were diagnosed in 27 institutions of the Italian Neuroblastoma Group and registered in the Registro Italiano Neuroblastoma (RINB) [17]. Of these, 182 (9.0%) met the diagnostic criteria for stage 4S, 10 of whom were excluded because of insufficient data, leaving 172 for analysis. RINB data were retrieved by reviewing patients' medical records. In accordance with Hsu et al. [18], presenting symptoms were defined as "minor" or "major", the latter being: (i) massive hepatomegaly, i.e., liver enlargement extending beyond the transversal umbilical line; (ii) dyspnea, i.e., tachypnea sometimes requiring O_2 supplementation; and (iii) organ dysfunctions, involving one or more of the following: gastro-intestinal tract, cardiovascular system, renal function, and coagulation pattern.

Diagnosis and diagnostic work-up

Tumor diagnosis was based on clinical and biochemical data, supported by adequate imaging, and usually confirmed by the histopathology report. The diagnostic work-up included bone marrow aspirates, local assays of urinary catecholamine metabolites, and serum LDH and ferritin. After the year 2000, histology was centrally reviewed according to the International Neuroblastoma Pathology Classification (INPC) criteria [19]. Biological characteristics of the tumors were assayed at the National Neuroblastoma Laboratory and included MYCN gene and chromosome 1p status, and DNA index [20]. The size of the primary tumor was retrospectively obtained from radiological reports, and the median diameter of 5 cm was taken to identify large masses. The presence of "surgical risk factors" [21], then named "image-defined risk factors" (IDRFs) by the International Neuroblastoma Risk Group (INRG) [22], were retrieved from surgical forms.

Treatment

Irradiation of enlarged livers was rarely performed. Resection of the primary tumor within the first few weeks after diagnosis (upfront resection) was encouraged when feasible with minimal risk. Late resection was carried out upon institutional decision. The term resection was defined as either the radical excision of the primary tumor or its excision with minimal residue. Excision that was less than complete, but greater than 50% was defined as partial resection, while biopsy was an operation aimed at obtaining a tumor fragment suitable for histological and biological examinations [14]. Chemotherapy was indicated in patients presenting or developing major symptoms: before the year 2000, it was administered in accordance with national protocols and consisted of 2–4 courses of various chemotherapeutic associations. After 2000, it consisted of the association of carboplatin and etoposide, according to an ad hoc SIOPEN protocol [23]. However, in the event of MYCN gene amplification, intensive upfront chemotherapy, followed by resection of the primary tumor and irradiation of the primary tumor site, was undertaken [24].

Statistical analyses

Descriptive statistics are reported as absolute frequencies and percentages for qualitative variables, and as median values with their related interquartile range (IQR) for quantitative variables. To compare proportions between groups, Pearson's Chi-square and Fisher's exact test, when appropriate, were applied. Progression-free survival (PFS) and overall survival (OS) were estimated by means of the Kaplan–Meier method, and differences between groups were assessed by means of the log-rank test. Survival estimates referred to the 5 years following diagnosis, and the related 95% confidence intervals (95% CI) were obtained by applying the Kalbfleisch and Prentice method [25]. Multivariable survival analysis, via Cox regression model, was limited to PFS, owing to the very low number of deaths recorded. All tests were two-tailed, and a P value < 0.05 was considered statistically significant. All analyses were performed by means of Stata Statistical Software (Release 13.1, Stata Corporation, College Station, TX, USA).

Results

Of 172 infants diagnosed with stage 4S neuroblastoma, 12 were excluded owing to early fatal disease progression ($n = 7$) or absence of an identifiable primary ($n = 5$), leaving 160 for analysis; 40 of these were diagnosed between 1994 and 1999 and 120 between 2000 and 2013. Of the 160 evaluable patients, 62 underwent upfront resection of the primary tumor and 98 did not. Late surgery was subsequently performed in 13 patients (2 of those who had undergone upfront resection, and 11 of those who had not).

Presenting features of the 160 infants evaluated for upfront surgery

Table 1 shows the main features of these 160 patients on diagnosis; 62 (38.8%) were scheduled for upfront resection, while the remaining 98 (61.2%) were scheduled for other kinds of treatment.

Gender and age

Male-to-female ratio was 1.1. Median age was 90 days, with 35.6% diagnosed within the first 2 months of life. No difference was observed between patients who underwent surgery and those who did not.

Symptoms

Sixteen patients (10.0%) were asymptomatic, as the tumor was detected in late pregnancy ($n = 2$), on post-natal screening ($n = 12$), or during follow-up of a neonatal adrenal mass ($n = 2$). Thirty-five patients presented with minor symptoms (21.9%); 109 (68.1%) presented with major symptoms: hepatomegaly in 83 (51.9%), dyspnea in 6 (3.8%), and the combination of both in 19 (11.9%). Patients who underwent surgery were more often asymptomatic (19.4% vs 4.1%) or had minor symptoms (29.0% vs 17.3%), and less frequently presented major symptoms (51.6% vs 78.6%).

Primary tumor site and size, and IDRFs

The primary tumor site was the adrenal in 106 infants (66.3%). The primary tumor size was recorded in 87 patients; in 28, the median diameter was greater than 5 cm. IDRFs were identified in 42 of the 98 patients who underwent this evaluation. Tumor size was similar in both groups, while patients who underwent upfront resection more often had an adrenal primary (83.9% vs 51.1%) and less frequently had IDRFs (21.6% vs 55.7%).

Metastatic sites

The liver was involved in 133 infants, bone marrow in 76, and skin in 16. Liver and skin involvement was more frequent in non-surgical patients (88.8% vs 74.2% and 14.3% vs 3.2%, respectively).

Histology and biology

Histology was centrally reviewed in 73 cases and deemed favorable in 67. MYCN gene was assayed in 147 tumors and found to be amplified in 12. Chromosome 1p was found to be deleted in 24 of 121 tumors tested, and the DNA index was di- or tetraploid in 36 of 106. The distribution of histological and biological features did not differ between surgical and non-surgical patients.

Treatment, clinical course, and outcome

Of the 62 patients who underwent upfront resection, 51 were assigned to observation, one of whom died of bleeding 6 days after surgery (Fig. 1). Nine of these 51 suffered disease progression 1–9 months after diagnosis (median 4): metastatic in 7 (one died 19 months after diagnosis) and combined in 2 (both alive); 5-year PFS and OS were, therefore, 82.0% and 96.1%, respectively. The remaining 11 of the 62 received chemotherapy; one suffered local disease progression at 2½ months and survived; PFS and OS were, therefore, 90.9% and 100%, respectively. Four patients suffered surgery-related complications: ischemic renal failure in two, intra-operative tumor rupture in one, and bilateral pleural effusion in one; all survived with appropriate treatment. Two of the 62 patients underwent a second, uncomplicated,

Table 1 Features on diagnosis of 160 infants with stage 4 s neuroblastoma evaluated for upfront resection of primary tumor

Features	All		Operated		Non-operated		*P*
	No	%	No	%	No	%	
Total	160	100	62	100	98	100	
M/F ratio	1.1		1.2		1.1		0.730
Median age, days (IQR)	90 (40–151)		97 (42–166)		76 (33–146)		0.214
Age							0.479
< 60 days	57	35.6	20	*32.3*	37	37.8	
≥ 60 days	103	64.4	42	67.7	61	62.2	
Symptoms							< 0.001
None	16	10.0	12	19.4	4	4.1	
Minor	35	21.9	18	29.0	17	17.3	
Major	109	68.1	32	51.6	77	78.6	–
Primary tumor sites							
Adrenal	106	66.3	52	83.9	54	55.1	< 0.001
Other	54	33.7	10	16.1	44	44.9	
Tumor size (87 evaluable)							0.954
< 5 cm	59	67.8	27	67.5	32	68.1	
≥ 5 cm	28	32.2	13	32.5	15	31.9	
IDRFs (98 evaluable)							0.001
Absent	56	57.1	29	78.4	27	44.3	
Present	42	42.9	8	21.6	34	55.7	
Metastatic sites							
Liver	133	83.1	46	74.2	87	88.8	0.016
Bone marrow	76	47.5	32	51.6	44	44.9	0.407
Skin	16	10.0	2	3.2	14	14.3	0.023
Histology (73 evaluable)							0.221
Favorable	67	91.8	31	96.9	36	87.8	
Unfavorable	6	8.2	1	3.1	5	12.2	
MYCN gene (147 evaluable)							0.226
Not amplified	135	91.8	58	95.1	77	89.5	
Amplified	12	8.2	3	4.9	9	10.5	
1p deletion (121 evaluable)							0.823
Absent	97	80.2	42	79.2	55	80.9	
Present	24	80.2	11	20.8	13	19.1	
DNA index (106 evaluable)							0.687
Triploid	70	66.0	34	68.0	36	64.3	
Diploid/tetraploid	36	34.0	16	32.0	20	35.7	

IQR interquartile range, *IDRFs* image-defined risk factors

operation 2 months after diagnosis, to remove a small tumor residue: both survived.

Of the 98 patients who did not undergo upfront resection, 53 were assigned to observation, 24 (45.3%) of whom suffered disease progression 1–30 months after diagnosis (median 4): metastatic in 14 (5 died), local in 2 (both alive), and combined in 8 (3 died, all with unfavorable biology); PFS and OS were, therefore, 54.7% and 84.9%, respectively. The remaining 45 received chemotherapy, which was complicated by toxic death in 3 cases. Ten of the 45 suffered disease progression 1–38 months after diagnosis (median 7): metastatic in 5 (3 died), local in 4 (no deaths), and combined in 1 (alive); PFS and OS were, therefore, 76.2% and 86.6%, respectively. In one of these 45 patients, a silastic patch was successfully applied to relieve abdominal tension. 11 out of the 98 patients underwent uncomplicated late resection of a residual tumor 3–22 months after diagnosis (median, 5); all but one survived.

In summary, 4 of 160 patients (2.5%) died of therapy-related complications, 44 (27.5%) suffered disease progression, which was only metastatic in 26 (16.3%; 9 deaths), only local in 7 (4.4%; no deaths), and combined in 11 (6.9%;

Groups	Upfront chemo		Toxic deaths	Disease progressions (deaths)				No. alive at 5 years	5-year PFS	5-year OS
				All	Metastatic	Local	Combined			
Operated	No	51	1	9 (1)	7 (1)	0	2 (0)	49	82.0%	96.1%
	Yes	11	0	1 (0)	0	1 (0)	0	11	90.9%	100%
	No/Yes	62	1	10 (1)	7 (1)	1 (0)	2 (0)	60	83.6%	96.8%
Non-operated	No	53	0	24 (8)	14 (5)	2 (0)	8 (3)	45	54.7%	84.9%
	Yes	45	3	10 (3)	5 (3)	4 (0)	1 (0)	39	76.2%	86.6%
	No/Yes	98	3	34 (11)	19 (8)	6 (0)	9 (3)	84	64.2%	85.7%
All	No/Yes	160	4	44 (12)	26 (9)	7 (0)	11 (3)	144	71.8%	90.0%

Fig. 1 Treatment, progressions, and outcomes of patients evaluable for upfront resection of primary tumor

3 deaths). The overall number of deaths was, therefore, 16 (10.0%); these occurred 6 days to 42 months (median, 10 months) after diagnosis. Twenty-nine of the 160 patients (18.1%) received no treatment at all and were alive at the last follow-up examination recorded. A total of 144 patients (90.0%) are alive after a follow-up of 60 months, 5-year PFS and OS being 71.8% and 90.0%, respectively.

Analysis of survival

The 5-year PFS of the 160 patients evaluated for upfront surgery was 71.8% (Fig. 2a). PFS was better in patients diagnosed in the first treatment era (76.9% *vs* 56.4%) and those presenting without symptoms or with minor symptoms in comparison with those with major symptoms (93.8% vs 82.9% vs 64.8%) (Table 2). In patients who underwent upfront resection, PFS was better in those diagnosed more recently (89.4% vs 64.3%) and those without IDRFs (89.7% vs 62.5%). In patients who did not undergo upfront resection, none of the presenting features was associated with better PFS. PFS was better in patients who underwent resection than in those who did not (83.6% vs 64.2%) (Fig. 2a). On comparing patients who underwent resection with those who did not, the features associated with better PFS in the former were: recent treatment era, older age, female gender, adrenal primary, absence of IDRFs, favorable histology, and normal *MYCN* status (Table 2).

Multivariable analysis confirmed the better PFS in patients who underwent either upfront primary resection (hazard ratio HR = 0.42) or chemotherapy (HR = 0.44), and the poorer survival of those presenting with major symptoms on diagnosis (HR = 2.4) (Table 3). In the upfront resection group, the better survival in the later treatment era (HR = 0.26) and the higher risk among those who presented with IDRFs (HR = 4.6) were also confirmed, though statistical significance was borderline (Supplementary Table 1). In non-surgical patients, a protective effect of upfront chemotherapy (HR = 0.44) was observed (Supplementary Table 1).

The 5-year OS of the 160 patients evaluated for upfront surgery was 90.0% (Fig. 2b). A better OS was associated with favorable histology, normal *MYCN* gene and chromosome 1p status, and triploid DNA index (Table 4). In patients who underwent resection, no presenting feature was associated with better OS, while in those who did not, OS was affected by unfavorable histological and biological features. OS was better in the former group than in the latter (96.8% vs 85.7%) (Fig. 2b). On comparing patients who underwent resection with those who did not, features associated with better OS were: adrenal site, large diameter of the primary and absence of IDRFs (Table 4). No multivariable analysis of OS was carried out, owing to the small number of deaths recorded.

Discussion

Back in 1996, Guglielmi et al. evaluated the effect of resecting the primary tumor in 97 Italian infants diagnosed with stage 4S neuroblastoma and found that those who underwent resection had no better outcome than those who did not [14]. Their conclusions, however, conflicted with those of several other reports [9–11, 13, 15]. This is a relevant

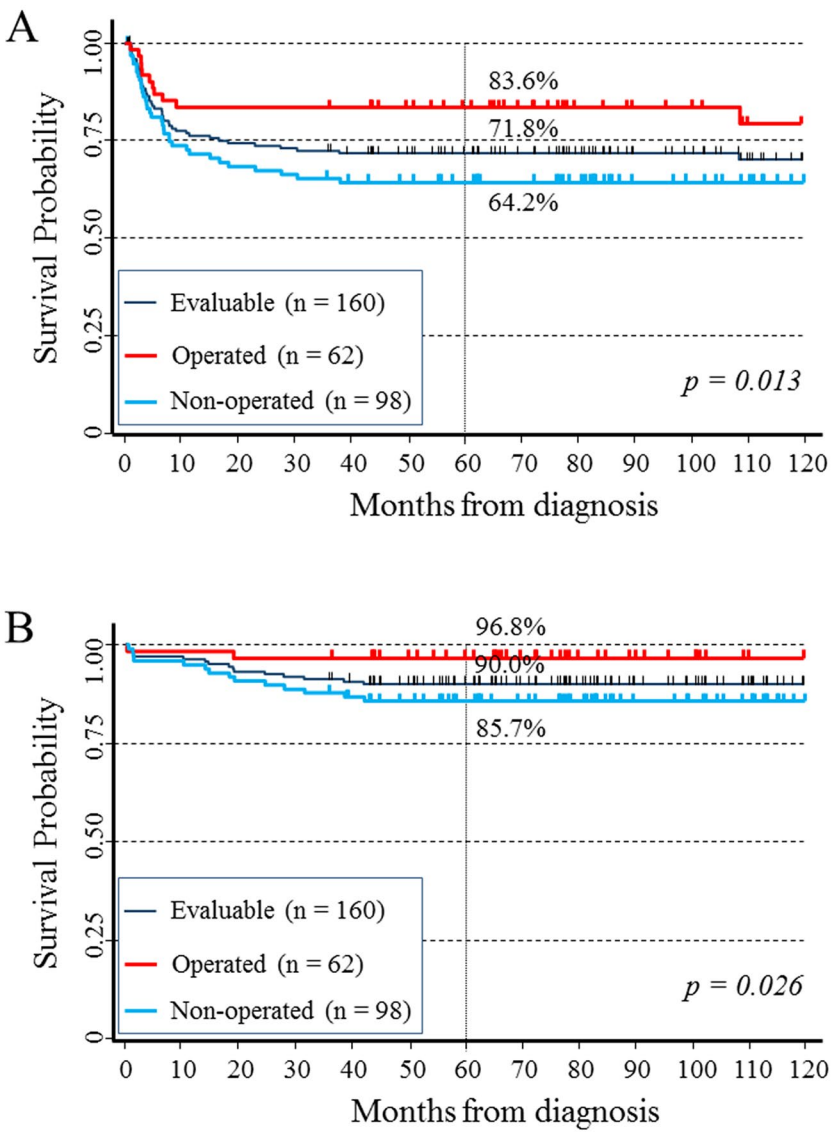

Fig. 2 **a** Progression-free survival of 160 patients with stage 4 s neuroblastoma evaluable for upfront resection of primary tumor. **b** Overall survival of 160 patients with stage 4S neuroblastoma evaluable for upfront resection of primary tumor

issue, as major surgery performed in small infants is not devoid of risk and should be considered with caution in the absence of clear advantage for the patient. With regard to this particular therapeutic aspect, we, therefore, analyzed an additional cohort of such patients diagnosed in Italy between 1994 and 2013. Of note, in the present study, infants who died of early progression or did not have an identifiable primary were excluded.

Without recognized treatment guidelines, physicians treating stage 4S neuroblastoma in the present study were recommended to resect the primary tumor, when this was deemed feasible without risk. This led to a 36% rate of upfront primary tumor resection in the whole series, which was greater than the 27% reported in Guglielmi's series [14] and may reflect an increased confidence of surgeons in operating on these patients. Upfront surgery was usually reserved for infants who presented with reassuring clinical conditions and tumor imaging, specifically, those without major symptoms or IDRFs, and those in whom the adrenal was the most frequent primary site. Nevertheless, a few patients underwent surgery despite having IDRFs. These did not suffer complications and survived. Indeed, it is well known that, in terms of surgical complexity, IDRFs are not all equivalent, and that some can be safely managed by expert surgeons, thereby avoiding chemotherapy in these young infants.

As in the previous study [14], PFS and OS rates in the present study were significantly better in patients who underwent upfront tumor resection, who accounted for approximately one-third of the cases ($n=62$, i.e., 36%). However, most of these patients were assigned to surgery owing to their favorable presenting features. Of note, 5 of the 62 suffered major surgery-related complications, one fatal. Whether this group of patients would have had the same

Table 2 Five-year PFS in relation to features on diagnosis of 160 stage 4 s neuroblastoma patients evaluated for upfront resection of primary tumor

Features	All patients (No = 160)				Operated (No = 62)				Non-operated (No = 98)				p^*
	No/PD	PFS%	95% CI	p	No/PD	PFS%	95% CI	p	No/PD	PFS%	95% CI	p	
Total	160/44	71.8	64.0–78.2	–	62/10	83.6	71.7–90.8	–	98/34	64.2	53.7–73.0	–	**0.013**
Treatment eras				**0.019**				**0.023**				0.210	
1994–1999	40/17	56.4	39.6–70.2		15/5	64.3	34.3–83.3		25/12	52.0	31.3–69.2		0.626
2000–2013	120/27	76.9	68.2–83.6		47/5	89.4	76.3–95.4		73/22	68.6	56.3–78.1		**0.011**
Age				0.462				0.589				0.657	
< 60 days	57/17	69.2	55.2–79.6		20/4	80.0	55.1–92.0		37/13	63.0	45.0–76.6		0.198
60–365 days	103/27	73.3	63.5–80.8		42/6	85.4	70.3–93.1		61/21	65.0	51.5–75.6		**0.035**
Gender				0.737				0.240				0.767	
Males	85/24	70.8	59.6–79.3		34/7	78.8	60.6–89.3		51/17	65.4	50.3–76.9		0.241
Females	75/20	73.0	61.3–81.6		28/3	89.3	70.4–96.4		47/17	63.0	47.5–75.2		**0.016**
Symptoms				**0.023**				0.424				0.187	
None	16/1	93.8	63.2–99.1		12/1	91.7	53.9–98.8		4/0	100	–		0.564
Minor	35/6	82.9	65.8–91.9		18/2	88.9	62.4–97.1		17/4	76.5	48.8–90.5		0.338
Major	109/37	64.8	54.9–73.1		32/7	77.4	58.4–88.5		77/30	59.5	47.5–69.7		0.108
Primary tumor sites				0.524				0.724				0.084	
Adrenal	106/31	69.9	60.1–77.8		52/8	84.3	71.1–91.8		54/23	55.9	41.4–68.1		**0.003**
Other	54/13	75.5	61.5–85.0		10/2	80.0	40.9–94.6		44/11	74.4	58.6–84.9		0.741
Tumor size				0.574				0.812				0.385	
< 5 cm	59/17	70.7	57.2–80.7		27/5	81.5	61.1–91.8		32/12	61.4	42.1–75.9		0.113
≥ 5 cm	28/10	64.3	43.8–78.9		13/2	84.6	51.2–95.9		15/8	46.7	21.2–68.8		0.053
IDRFs				0.073				**0.044**				0.806	
Absent	56/14	75.0	61.5–84.4		29/3	89.7	71.3–96.5		27/11	59.3	38.6–75.0		**0.012**
Present	42/17	59.5	43.2–72.6		8/3	62.5	22.9–86.1		34/14	58.8	40.6–73.2		0.939
Histology				0.615				0.752				0.893	
Unfavorable	6/2	66.7	19.5–90.4		1/0	100	-		5/2	60.0	12.6–88.2		0.502
Favorable	67/14	78.8	66.9–86.9		31/3	90.3	72.9–96.8		36/11	68.6	50.5–81.2		**0.036**
MYCN status				0.443				0.447				0.871	
Normal[§]	135/37	72.0	63.5–78.8		58/9	84.2	71.9–91.5		77/28	62.7	50.7–72.5		**0.009**
Amplified	12/5	58.3	27.0–80.0		3/1	66.7	5.4–94.5		9/4	55.6	20.4–80.5		0.838
1p status				0.290				0.547				0.366	
Normal	97/26	72.6	62.4–80.4		42/7	82.9	67.5–91.5		55/19	64.8	50.5–75.9		0.065
Deleted	24/10	58.3	36.5–75.0		11/3	72.7	37.1–90.3		13/7	46.2	19.2–69.6		0.258
DNA Index				0.808				0.594				0.923	
Triploid	70/20	71.4	59.3–80.5		34/6	82.4	64.9–91.7		36/14	61.1	43.4–74.8		0.055
Diploid/Tetraploid	36/10	72.2	54.5–84.0		16/2	87.5	58.6–96.7		20/8	60.0	35.7–77.6		0.072

Bold p value indicates a statistically significant value < 0.05

No/PD number of patients/number of progressions, *PFS%* progression-free survival per 100 at 5 years after diagnosis, *95% CI* 95% confidence interval, *p p* value estimated by the log-rank test. *p** comparison between patients with no elective surgery and patients with tumor resection

excellent outcome if they had not undergone primary resection remains uncertain.

A larger group ($n = 98$, i.e., 57%) was made up of patients judged unsuitable for primary tumor resection. Interestingly, both patients presenting with reassuring conditions who were assigned to observation and those who underwent upfront chemotherapy owing to the presence of major symptoms had similar OS (84.9% and 86.6%), suggesting that upfront chemotherapy may have improved the outcome of patients presenting with ominous clinical features. A similar favorable effect was reported in a recent COG study, in which stage 4S infants received pre-emptive chemotherapy owing to their evolving symptoms and/or unfavorable biological features [16]. In the present study, unfavorable biology in non-surgical patients was associated with a higher risk of disease progression and death. In accordance with the

Table 3 Five-year PFS of 160 stage 4 s neuroblastoma patients by features on diagnosis and upfront therapies

Patient features	HR	95% CI	*p*
Upfront primary tumor resection	0.42	0.20–0.88	**0.022**
Upfront chemotherapy	0.44	0.22–0.89	**0.021**
Treatment era 2000–2013	0.72	0.37–1.4	0.333
Age on diagnosis ≥ 60 days	0.76	0.41–1.4	0.392
Female gender	0.86	0.48–1.6	0.631
Presence of major symptoms	2.4	1.1–5.6	**0.036**
Adrenal primary site	1.4	0.72–2.8	0.311
Primary tumor diameter < 5 cm	0.77	0.35–1.7	0.521
IDRFs	1.8	0.81–4.1	0.143
Favorable histology	0.88	0.20–4.0	0.873
MYCN gene amplification	1.6	0.63–4.3	0.310
Chromosome 1p deletion	1.5	0.73–3.2	0.260
Diploid/tetraploid DNA index	0.99	0.46–2.1	0.971

Bold *p* value indicates a statistically significant value < 0.05

IDRFs image-defined risk factors, *HR* hazard ratio evaluated by multivariable Cox regression analysis, *95% CI* 95% confidence interval, *p p* value

SIOPEN recommendations [26], we emphasize the importance of obtaining information on these features at the onset, as an intensive approach has proved effective in patients with unfavorable biology [23].

It is of interest that 18 of the total 44 cases of tumor progression involved the primary site and occurred in 3/62 surgical and 15/98 non-surgical patients. There were three instances of fatal local progression, all of which occurred among non-surgical patients who had unfavorable biological features. As previously reported [14], we may conclude that the primary tumor site was involved in a minority of cases and that progression at this site only occasionally contributed to death.

Delayed surgery to remove a residual tumor was carried out in 13 patients, 11 of whom had not previously undergone resection. None of these patients experienced post-operative complications and only one died.

This study had some limitations: (i) the retrospective design of the study, which spanned 19 years, (ii) the small sample size of each group/subgroup of patient features, (iii) the small number of disease-related or therapy-related events and deaths observed, and (iv) the high number of statistical comparisons, which engenders a risk of multiple testing bias. Nevertheless, our data clearly show that the outcome was better in patients diagnosed in the second treatment era (92.5% vs 82.5%). As presenting features were comparable in the two periods, the better result of those more recently diagnosed could be attributed to a combination of factors: (i) a more-refined management strategy, resulting from participation in a large international study, (ii) the use of optimal chemotherapy (the carboplatin–etoposide association) rather than previous less effective combinations [10], (iii) the success of adopting an aggressive treatment in patients with amplified MYCN gene [24], and (iv) the increasing tendency to treat critical patients in intensive care units for severely ill neonates. On the basis of these considerations, future studies on this topic should focus on patients treated in more recent eras.

Although patients who underwent primary resection had a better outcome than those who did not, this result cannot be entirely or definitely attributed to surgery, since patients were selected for resection on the basis of their favorable features. A thoroughly preoperative multidisciplinary discussion is mandatory, as operating on these young patients is not devoid of risk, and patients may require other kinds of upfront treatment rather than surgery.

Our long-lasting retrospective experience revealed that: (a) small primaries were generally either kept under observation or easily and safely resected, leaving little or no space for other treatments based on tumor biology; (b) primaries in which upfront surgery was hazardous and could not guarantee complete resection benefited from upfront chemotherapy, which was usually preceded by adequate tumor biopsy to evaluate biology; (c) delayed surgery was carried out only on a few patients, on the basis of local staff decision; (d) in those few patients who presented with tumor progression despite chemotherapy (4.7% in our series), surgery played a minimal role, being performed only on those who had life-threatening symptoms. These latter patients may benefit from emergency surgical procedures to support vital functions, such as temporary positioning of an abdominal silastic patch [27], intra-arterial liver chemoembolization [28], and liver transplantation [29].

To reduce surgical morbidity and chemotherapy toxicity, and to provide uniformity in the treatment of low and intermediate neuroblastoma (including stage 4S), an ongoing SIOPEN study has developed a therapeutic algorithm based on tumor imaging and biological features [26, 30].

A recent systematic review on the outcome of stage 4S neuroblastoma confirmed that significant mortality is still observed in these patients, and that those with *MYCN* gene amplification and 1p/11q deletion have a dismal outcome [31]. The authors concluded that patients amenable to conservative management or surgery to excise the primary tumor have the best prognosis.

Although the question of whether to operate or not at disease onset is still unsolved, this study confirms the importance of obtaining enough adequate tumor tissue to enable histology and biology studies to properly address treatment, to achieve the best possible outcome.

Table 4 Five-year OS in relation to features on diagnosis in 160 stage 4 s neuroblastoma patients evaluated for upfront resection of primary tumor

Features	All (No = 160)				Operated (No = 62)				Non-operated (No = 98)				p^*
	No/D	OS%	95% CI	*p*	No/D	OS%	95% CI	*p*	No/D	OS%	95%CI	*p*	
Total	160/16	90.0	84.2–93.7	–	62/2	96.8	87.7–99.2	–	98/14	85.7	77.0–91.3	–	**0.026**
Treatment eras				0.072				0.379				0.129	
1994–1999	40/7	82.5	66.7–91.3		15/1	93.3	61.3–99.0		25/6	76.0	54.2–88.4		0.191
2000—2013	120/9	92.5	86.1–96.0		47/1	97.9	85.8–99.7		73/8	89.0	79.2–94.4		0.075
Age				0.479				0.598				0.663	
< 60 days	57/7	87.7	76.0–94.0		20/1	95.0	69.5–99.3		37/6	83.8	67.4–92.4		0.222
60–365 days	103/9	91.3	83.9–95.4		42/1	97.6	84.3–99.7		61/8	86.9	75.5–93.2		0.064
Gender				0.190				0.196				0.338	
Males	85/11	87.0	77.8–92.6		34/2	94.1	78.5–98.5		51/9	82.4	68.8–90.4		0.126
Females	75/5	93.3	84.7–97.2		28/0	100	–		47/5	89.3	76.2–95.4		0.077
Symptoms				0.070				0.386				0.371	
None	16/0	100	–		12/0	100	–		4/0	100	–		NA
Minor	35/1	97.1	81.4–99.6		18/0	100	–		17/1	94.1	65.0–99.2		0.304
Major	109/15	86.2	78.2–91.5		32/2	93.8	77.3–98.4		77/13	83.1	72.7–89.8		0.155
Primary tumor sites				0.460				0.533				0.209	
Adrenal	106/12	88.7	80.9–93.4		52/2	96.2	85.5–99.0		54/10	81.4	68.3–89.6		**0.019**
Other site	54/4	92.6	81.5–97.2		10/0	100	–		44/4	90.9	77.6–96.5		0.332
Tumor size				0.314				0.488				0.170	
< 5 cm	59/6	89.8	78.8–95.3		27/1	96.3	76.5–99.5		32/5	84.4	66.5–93.2		0.134
≥ 5 cm	28/5	82.1	62.3–92.2		13/0	100	–		15/5	66.7	37.5–84.6		**0.025**
IDRFs				0.401				NA				0.967	
Absent	56/4	92.9	82.1–97.3		29/0	100	–		27/4	85.2	65.2–94.2		**0.033**
Present	42/5	88.1	73.7–94.9		8/0	100	–		34/5	85.3	68.2–93.6		0.263
Histology				**0.009**				0.858				**0.019**	
Unfavorable	6/2	66.7	19.5–90.4		1/0	100	–		5/2	60.0	12.6–88.2		0.502
Favorable	67/3	95.5	86.8–98.5		31/1	96.8	79.2–99.5		36/2	94.4	79.6–98.6		0.636
MYCN status				**0.006**				0.747				**0.009**	
Normal[§]	135/11	91.8	85.7–95.4		58/2	96.6	86.9–99.1		77/9	88.2	78.6–93.7		0.087
Amplified	12/4	66.7	33.7–86.0		3/0	100	–		9/4	55.6	20.4–80.5		0.201
1p status				**0.002**				0.313				**0.002**	
Normal	97/7	92.7	85.3–96.5		42/1	97.6	84.3–99.7		55/6	89.0	77.1–94.9		0.115
Deleted	24/7	70.8	48.4–84.9		11/1	90.9	50.8–98.7		13/6	53.9	24.8–76.0		0.059
DNA Index				**< 0.001**				0.145				**0.004**	
Triploid	70/1	98.6	90.3–99.8		34/0	100	–		36/1	97.2	81.9–99.6		0.331
Diploid/Tetraploid	36/7	80.5	63.3–90.2		16/1	93.8	63.2–99.1		20/6	70.0	45.1–85.3		0.082

Bold *p* value indicates a statistically significant value < 0.05

No/D number of patients/number of deaths, *OS%* overall survival per 100 at 5 years after diagnosis, *95% CI* 95% confidence interval, *NA* not available, *p** comparison between patients with no elective surgery and patients with tumor resection

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Author contributions SA, GM, AG, MC, and BDB: study design, data interpretation, manuscript drafting, and main revision. IB, ARG, and GE: data analysis and manuscript drafting. SP: statistical analysis, manuscript drafting, and revision. IP: review of patients' charts and data analysis. CG: review of patients' imaging and manuscript drafting. ARS: review of patients' histology and manuscript drafting. AI, PDI, AMF, ML, UC, and ET: provided patient data, and edited and revised the manuscript;

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval The RINB structure and protocol were approved by all the ethics committees of each participating center as a retrospective and prospective observational study. The RINB database is located at the secure Italian Inter-University Consortium CINECA headquarters in Italy, which is 9001:2015 and 27,001:2013 certified. It can be accessed only by authorized users. This retrospective cohort study was conducted in accordance with the ethical standards of the institutional and national research committees and with the 1964 Helsinki Declaration and its later amendments.

Informed consent To be enrolled in the RINB, an informed consent form had to be signed by the patient's parents or guardians. For this reason, no specific further consent for this retrospective study needed to be sought.

References

1. D'Angio GJ, Evans AE, Koop C (1971) Special pattern of widespread neuroblastoma with a favorable prognosis. Lancet 1:1046–1049. https://doi.org/10.1016/s0140-6736(71)91606-0
2. Brodeur GM, Pritchard J, Berthold F et al (1993) Revisions of the international criteria for neuroblastoma diagnosis, staging, and response to treatment. J Clin Oncol 11:1466–1477. https://doi.org/10.1200/JCO.1993.11.8.1466
3. Brodeur GM (2018) Spontaneous regression of neuroblastoma. Cell Tissue Res. 372:277–286. https://doi.org/10.1007/s00441-017-2761-2
4. Evans AE, Chatten J, D'Angio GJ, Gerson JM, Robinson J, Schnaufer L (1980) A review of 17 IV-S neuroblastoma patients at the Children's Hospital of Philadelphia. Cancer 45:833–839. https://doi.org/10.1002/1097-0142(19800301)45:5%3c833::aid-cncr2820450502%3e3.0.co;2-u
5. Stokes SH, Thomas PR, Perez CA, Vietti TJ (1984) Stage IV-S neuroblastoma: results with definitive therapy. Cancer 53:2083–2086. https://doi.org/10.1002/1097-0142(19840515)53:10%3c2083::aid-cncr2820531014%3e3.0.co;2-s
6. De Bernardi B, Pianca C, Boni L, Brisigotti M, Carli M, Bagnulo S, Corciulo P, Mancini A, De Laurentis C, Di Tullio MT et al (1992) Disseminated neuroblastoma (stage IV and IV-S) in the first year of life. Cancer 70:1625–1633. https://doi.org/10.1002/1097-0142(19920915)70:6%3c1625::aid-cncr2820700631%3e3.0.co;2-6
7. Blatt J, Deutsch M, Wollman MR (1987) Results of therapy in stage IV-s neuroblastoma with massive hepatomegaly. Int J Radiat Oncol Biol Phys 13:1467–1471. https://doi.org/10.1016/0360-3016(87)90312-9
8. Katzenstein HM, Bowman LC, Brodeur GM, Thorner PS, Joshi VV, Smith EI, Look AT, Rowe ST, Nash MB, Holbrook T, Alvarado C, Rao PV, Castleberry RP, Cohn SL (1998) Prognostic significance of age, MYCN oncogene amplification, tumor cell ploidy, and histology in 110 infants with stage D(S) neuroblastoma: the Pediatric Oncology Group experience. J Clin Oncol 6:2007–2017. https://doi.org/10.1200/JCO.1998.16.6.2007
9. Nickerson HJ, Matthay KK, Seeger RC, Brodeur GM, Shimada H, Perez C, Atkinson JB, Selch M, Gerbing RB, Stram DO, Lukens J (2000) Favorable biology and outcome of stage IV-S neuroblastoma with supportive care or minimal therapy: a Children's Cancer Group study. J Clin Oncol 18:477–486. https://doi.org/10.1200/JCO.2000.18.3.477
10. Schleiermacher G, Rubie H, Hartmann O, Bergeron C, Chastagner P, Mechinaud F, Michon J (2003) Neuroblastoma Study Group of the French Society of Paediatric Oncology Treatment of stage 4S neuroblastoma—report of 10 years' experience of the French Society of Paediatric Oncology (SFOP). Br J Cancer 89:470–476. https://doi.org/10.1038/sj.bjc.6601154
11. Taggart DR, London WB, Schmidt ML, DuBois SG, Monclair TF, Nakagawara A, De Bernardi B, Ambros PF, Pearson AD, Cohn SL, Matthay KK (2011) Prognostic value of the stage 4S metastatic pattern and tumor biology in patients with metastatic neuroblastoma diagnosed between birth and 18 months of age. J Clin Oncol 29:4358–4364. https://doi.org/10.1200/JCO.2011.35.9570
12. De Bernardi B, Di Cataldo A, Garaventa A, Massirio P, Viscardi E, Podda MG, Castellano A, D'Angelo P, Tirtei E, Melchionda F, Vetrella S, De Leonardis F, D'Ippolito C, Tondo A, Nonnis A, Erminio G, Gigliotti AR, Mazzocco K, Haupt R (2019) Stage 4s neuroblastoma: features, management and outcome of 268 cases from the Italian Neuroblastoma Registry. Ital J Pediatr 45:8–23. https://doi.org/10.1186/s13052-018-0599-1
13. Martinez DA, King DR, Ginn-Pease ME, Haase GM, Wiener ES (1992) Resection of the primary tumor is appropriate for children with stage IV-S neuroblastoma: an analysis of 37 patients. J Pediatr Surg 27:1016–1020. https://doi.org/10.1016/0022-3468(92)90549-m
14. Guglielmi M, De Bernardi B, Rizzo A, Federici S, Boglino C, Siracusa F, Leggio A, Cozzi F, Cecchetto G, Musi L, Bardini T, Fagnani AM, Bartoli GC, Pampaloni A, Rogers D, Conte M, Milanaccio C, Bruzzi P (1996) Resection of primary tumor at diagnosis in stage IV-S neuroblastoma: does it affect the clinical course? J Clin Oncol 14:1537–1544. https://doi.org/10.1200/JCO.1996.14.5.1537
15. Berthold F, Harms D, Lampert F, Niethammer D, Zieschang J (1990) Risk factors in neuroblastoma of infants. Contrib Oncol 41:101–117. https://doi.org/10.1159/000419225
16. Twist CJ, Naranjo A, Schmidt ML, Tenney SC, Cohn SL, Meany HJ, Mattei P, Adkins ES, Shimada H, London WB, Park JR, Matthay KK, Maris JM (2019) Defining risk factors for chemotherapeutic intervention in infants with stage 4S neuroblastoma: a report from Children's Oncology Group Study ANBL0531. J Clin Oncol 37:115–124. https://doi.org/10.1200/JCO.18.00419
17. Haupt R, Garaventa A, Gambini C, Parodi S, Cangemi G, Casale F, Viscardi E, Bianchi M, Prete A, Jenkner A, Luksch R, Di Cataldo A, Favre C, D'Angelo P, Zanazzo GA, Arcamone G, Izzi GC, Gigliotti AR, Pastore G, De Bernardi B (2010) lmproved survival of children with neuroblastoma between 1979 and 2005: a report of the Italian Neuroblastoma Registry. J Clin Oncol 28:2331–2338. https://doi.org/10.1200/JCO.2009.24.8351
18. Hsu LL, Evans AE, D'Angio GJ (1996) Hepatomegaly in neuroblastoma stage 4s: criteria for treatment of the vulnerable neonate. Med Pediatr Oncol 27:521–528. https://doi.org/10.1002/(SICI)1096-911X(199612)27:6%3c521::AID-MPO3%3e3.0.CO;2-N
19. Shimada H, Ambros IM, Dehner LP, Hata J, Joshi VV, Roald B, Stram DO, Gerbing RB, Lukens JN, Matthay KK, Castleberry RP (1999) The International neuroblastoma pathology classification (the Shimada system). Cancer 86:364–372. https://doi.

org/10.1002/(SICI)1097-0142(19990715)86:2%3c364::AID-CNCR21%3e3.0.CO;2-7
20. Scaruffi P, Parodi S, Mazzocco K, Defferrari R, Fontana V, Bonassi S, Tonini GP (2004) Detection of MYCN amplification and chromosome 1p36 loss in neuroblastoma by cDNA microarray comparative genomic hybridization. Mol Diagn 8:93–100. https://doi.org/10.1007/BF03260051
21. Cecchetto G, Mosseri V, De Bernardi B, Helardot P, Monclair T, Costa E, Horcher E, Neuenschwander S, Tomà P, Rizzo A, Michon J, Holmes K (2005) Surgical risk factors in primary surgery for localized neuroblastoma: the LNESG1 Study of the European International Society of Pediatric Oncology Neuroblastoma Group. J Clin Oncol 23:8483–8489. https://doi.org/10.1200/JCO.2005.02.4661
22. Monclair T, Brodeur GM, Ambros PF, Brisse HJ, Cecchetto G, Holmes K, Kaneko M, London WB, Matthay KK, Nuchtern JG, von Schweinitz D, Simon T, Cohn SL, Pearson AD, INRG Task Force (2009) The International Neuroblastoma Risk Group (INRG) staging system: an INRG Task Force report. J Clin Oncol 27:298–303. https://doi.org/10.1200/JCO.2008.16.6876
23. De Bernardi B, Gerrard M, Boni L, Rubie H, Cañete A, Di Cataldo A, Castel V, Forjaz de Lacerda A, Ladenstein R, Ruud E, Brichard B, Couturier J, Ellershaw C, Munzer C, Bruzzi P, Michon J, Pearson AD (2009) Excellent outcome with reduced treatment for infants with disseminated neuroblastoma without MYCN amplification. J Clin Oncol 27:1034–1040. https://doi.org/10.1200/JCO.2008.17.5877
24. Canete A, Gerrard M, Rubie H, Castel V, Di Cataldo A, Munzer C, Ladenstein R, Brichard B, Bermúdez JD, Couturier J, de Bernardi B, Pearson AJ, Michon J (2009) Poor survival for infants with MYCN-amplified metastatic neuroblastoma despite intensified treatment: the International Society of Paediatric Oncology European Neuroblastoma Experience. J Clin Oncol 27:1014–1019. https://doi.org/10.1200/JCO.2007.14.5839
25. Hosmer DW, Lemeshow S (1999) Applied survival analysis—regression modeling of time to event data. Wiley, New York
26. European Low and Intermediate Neuroblastoma: a SIOPEN Study version 3.0, 30 May 2011 https://www.docvadis.fr/files/all/JI3RO1LMf9PWrlqN9k7X7w/neuroblastome_low_intermediate_risk_lines_protocol_version_4_2_2_janvier_2013.pdf.
27. Keene DJ, Minford J, Craigie RJ, Humphrey G, Bruce J (2001) Laparostomy closure in stage 4S neuroblastoma. J Pediatr Surg 46:1–4. https://doi.org/10.1016/j.jpedsurg.2010.08.064
28. Weintraub M, Bloom AI, Gross E, Revel-Vilk S, Shahroor S, Koplewitz BZ, Freeman AI (2004) Successful treatment of progressive stage 4s hepatic neuroblastoma in a neonate with intra-arterial chemoembolization. Pediatr Blood Cancer 43:148–151. https://doi.org/10.1002/pbc.20080
29. Holsten T, Schuster T, Grabhorn E, Hero B, Frühwald MC (2017) Liver transplantation as a potentially life-saving measure in neuroblastoma stage 4S. Pediatr Hematol Oncol 34:17–23. https://doi.org/10.1080/08880018.2016.1266535
30. Schleiermacher G, Michon J, Ribeiro A, Pierron G, Mosseri V, Rubie H, Munzer C, Bénard J, Auger N, Combaret V, Janoueix-Lerosey I, Pearson A, Tweddle DA, Bown N, Gerrard M, Wheeler K, Noguera R, Villamon E, Cañete A, Castel V, Marques B, de Lacerda A, Tonini GP, Mazzocco K, Defferrari R, De Bernardi B, Di Cataldo A, van Roy N, Brichard B, Ladenstein R, Ambros I, Ambros P, Beiske K, Delattre O, Couturier J (2011) Segmental chromosomal alterations lead to a higher risk of relapse in infants with MYCN-non-amplified localized unresectable/disseminated neuroblastoma (a SIOPEN collaborative study). Br J Cancer 107:1418–1422. https://doi.org/10.1038/bjc.2011.472
31. Raitio A, Rice MJ, Mullassery D, Losty PD (2020) Stage 4S neuroblastoma: what are the outcomes? A systematic review of published studies. Eur J Pediatr Surg. https://doi.org/10.1055/s-0040-1716836.10.1055/s-0040-1716836 (**published online ahead of print, 2020 Sep 15**)