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CLASSIFICATION AND SELECTION OF IMAGE DEFINED RISK FACTORS IN NEUROBLASTOMA

DISSERTATION

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I. OVERVIEW OF THE AVAILABLE LITERATURE

Neuroblastoma (NBL) is the most common embryonal tumor and the third most frequent malignant tumor in children (after leukaemia and brain malignancies). NBL is the most frequent extracranial tumor before 18 years of age as it makes up 8-10% of pediatric neoplasms. It accounts for about 15% of childhood cancer deaths. It stems from the primitive cells of the sympathetic nervous system and of the adrenal medulla. Between 25% and 35% of cases manifest before the first year, while 75% manifest by the fourth. 90% of cases are diagnosed before 6 years of age. Only 3% of cases present after 10 years of age. Mean age of diagnosis is 2 years. Neuroblastoma yearly morbidity varies between 5-10 per million children. There are no racial differences in frequency, though such are described in tumor biology – African-Americans are more likely to have a fatal outcome. Mean age of diagnosis is about 18-22 months. Approximately 40-70% of patients have metastatic disease at diagnosis. Survivability is highly dependant on the age at which which diagnosis is established and the stage of disease – children below 1 year of age and at an early stage have the best prognosis. NBLs have a variable course – some behave aggressively, while others (typically in infancy) can spontaneously regress – a rather unique characteristic.

NBL is most frequently located in the abdomen – 65-70% of cases. The adrenal glands make up 35% of all cases as the most common site of origin, while the extraadrenal retroperitoneum (the paraspinal ganglia and, to a lesser degree, the organ of Zuckerkandl) primaries comprise 30% of cases. Mediastinal cases are 15%, cervical – 4%, spinal ganglia – 8%, pelvis – 2%, and olfactory apparatus – 1%. Multiple primary tumors can occur in up to 2% of cases, potentially with varying degrees of differentiation.

Newborns and infants can be affected (congenital and neonatal neuroblastoma). Neuroblastoma is the most common malignancy in the first month of life. Neonatal NBL is of adrenal origin. It has a very good prognosis, and despite metastatic spread present at diagnosis in almost half of patients, survival rates are over 90%. Fetal NBL can be detected on antenatal ultrasonography or magnetic resonance tomography as
early as 19 weeks (mean age of discovery 36 weeks gestation) – it is adrenal in origin in 90% of cases. Fetal neuroblastoma also has a very good prognosis, treatment is conservative.

**NEUROBLASTOMA STAGING SYSTEMS**

The treatment and prognosis of NBL are determined by the disease’s stage at diagnosis. Currently there are two methods of staging – one based on post-operative patients (INSS), and one for pre-treatment patients (INRGSS).

The **International Neuroblastoma Staging System (INSS)** is for post-operative patients and its main purpose is prognosis of outcome. It was established in 1986 and revised in 1988.

**Stage 1**
- localised tumor with complete gross excision with or without microscopic residual disease
- contralateral and representative ipsilateral regional lymph nodes negative for disease (nodes attached to and removed with primary tumor may be positive)

**Stage 2a**
- localised tumor with incomplete gross excision
- ipsilateral and contralateral nodes negative for tumor

**Stage 2b**
- localised tumor with complete or incomplete resection
- positive ipsilateral (non-adherent) nodes
- contralateral nodes negative for tumor

**Stage 3**
- unresectable lateral tumor that crosses the midline or
- localised tumor with contralateral regional lymph node involvement
- midline tumour with bilateral extension by infiltration or by lymph node involvement

**Stage 4S ("special")**
Stage 4
  o distant metastases not fulfilling stage 4S

In 2008 the International Neuroblastoma Risk Group (INRG) introduced a clinical system for pretherapeutic (including preoperative) staging, based on imaging-determined risk factors – the **International Neuroblastoma Risk Group Staging System (INRGSS)**. Two stages of localized disease are identified – L1 and L2, as well as two stages of metastatic disease – M and MS. INRGSS evaluates whether the primary tumor is affected by one or more of nearly 20 specific Image Defined Risk Factors (IDRF).

**Stage L1**
  o localised tumors confined to one body cavity and not involving IDRFs

**Stage L2**
  o locoregional tumors involving one or more IDRFs

**Stage M**
  o distant (remote) metastases (i.e. excludes metastases to local lymph node groups)
  o excludes stage MS

**Stage MS**
  o metastases in patients <18 months (some centres <12 months) confined to skin, liver and/or bone marrow
Table 1. *Image Defined Risk Factors (IDRFs) in neuroblastic tumors*

<table>
<thead>
<tr>
<th>Ipsilateral tumor extension within two bodily compartments:</th>
<th>Neck-chest, chest-abdomen, abdomen-pelvis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neck:</td>
<td>Tumor encasing carotid and/or vertebral artery and/or internal jugular vein</td>
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<tr>
<td></td>
<td>Tumor extending to base of skull</td>
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<tr>
<td></td>
<td>Tumor compressing the trachea</td>
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<tr>
<td>Cervico-thoracic junction:</td>
<td>Tumor encasing brachial plexus roots</td>
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<tr>
<td></td>
<td>Tumor encasing subclavian vessels and/or vertebral and/or carotid artery</td>
</tr>
<tr>
<td></td>
<td>Tumor compressing the trachea</td>
</tr>
<tr>
<td>Thorax:</td>
<td>Tumor encasing the aorta and/or major branches</td>
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<tr>
<td></td>
<td>Tumor compressing the trachea and/or principal bronchi</td>
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<tr>
<td></td>
<td>Lower mediastinal tumor, infiltrating the costo-vertebral junction between T9 and T12</td>
</tr>
<tr>
<td>Thoraco-abdominal:</td>
<td>Tumor encasing the aorta and/or vena cava</td>
</tr>
<tr>
<td>Abdomen/pelvis:</td>
<td>Tumor infiltrating the porta hepatis and/or the hepatoduodenal ligament</td>
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<tr>
<td></td>
<td>Tumor encasing branches of the superior mesenteric artery at the mesenteric root</td>
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<td></td>
<td>Tumor encasing the origin of the coeliac axis, and/or of the superior mesenteric artery</td>
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<tr>
<td></td>
<td>Tumor invading one or both renal pedicles</td>
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<tr>
<td></td>
<td>Tumor encasing the aorta and/or vena cava</td>
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<tr>
<td></td>
<td>Tumor encasing the iliac vessels</td>
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<td></td>
<td>Pelvic tumor crossing the sciatic notch</td>
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<tr>
<td>Intraspinal tumor extension:</td>
<td>Intraspinal tumor extension (any level) provided that more than one-third of spinal canal in axial plane is invaded, the perimedullary leptomeningeal spaces are not visible, or the spinal cord intensity is abnormal</td>
</tr>
<tr>
<td>Infiltration of adjacent organs/structures:</td>
<td>Pericardium, diaphragm, kidney, liver, duodeno-pancreatic block and mesentery</td>
</tr>
<tr>
<td>Conditions to be recorded, but NOT considered IDRFs</td>
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<td>--------------------------------------------------</td>
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<tr>
<td>Multifocal primary tumors</td>
<td></td>
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<tr>
<td>Intraspinal tumors (with or without symptoms of spinal cord compression)</td>
<td></td>
</tr>
<tr>
<td>Pleural effusion (with or without malignant cells)</td>
<td></td>
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<tr>
<td>Ascites (with or without malignant cells)</td>
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</tr>
</tbody>
</table>

INRGSS and INSS are used in unison, complementing each other.

II. AIM OF THE STUDY
To classify the image defined risk factors (IDRFs) in neuroblastoma in accordance to their potential effect on survivability for the purpose of augmenting imaging descriptions, bettering prognosis establishment and treatment.

III. MATERIAL
This study constitutes a retrospective analysis of 42 patients – 16 boys and 26 girls, who have all been admitted in the UMHAT “Saint Marina” during the span 2000 – 2017. All patients were restaged in accordance to INSS and INRGSS criteria by a single radiologist using the imaging studies carried out at diagnosis. IDRF status was recorded for all patients. Event-Free Survival and Overall Survival times were established for all patients.

IV. UTILIZED IMAGING METHODS
Computed tomography images were acquired using a 64-slice Dual Source computed tomography machine Siemens Definition. Magnetic resonance images were acquired with a 1,5T GE Signa and a 3T magnetic resonance tomography machine Siemens Magnetom Verio. When archived DICOM images were unavailable, imaging information was gathered from available medical history.
V. UTILIZED STATISTICAL METHODS

The following statistical methods were used:

1. Descriptive statistical analysis.
2. Student’s t-test.
3. Dispersion analysis.
4. Survival analysis.
5. Logistic regression analysis.

RESULTS AND DISCUSSION

The most frequent IDRF in the studied sample was identified to be the infiltration of the renal pedicle (uni- / bilateral). This is a natural consequence of the body compartment distribution of neuroblastoma’s preferred organs of origin. NBL develops most commonly in the abdomen (65-70%), arising primarily from the adrenal glands (up to 35% of all neuroblastoma cases). The frequency with which this IDRF is idetified is augmented by the lowered imaging criteria for establishing its presence – mere contact between the primary mass and a renal vessel is sufficient. This is necessitated by the high risk of renal infarct / nephrectomy during attempts to separate the tumor from the renal vessels.

The second most frequent IDRF in the sample was found to be intraspinal growth and compression. The anatomical predilections of neuroblastoma explain this finding as well – the tumor arises from the paravertebral sympathetic chain (which is the most frequent organ of origin in the thorax) and can easily grow into the vertebral canal through the neuroforamina.

The place of third most frequent IDRF was shared among two – encasement of the celiac trunk / superior mesenteric vein and encasement of the abdominal aorta / inferior vena cava. Once again this can be explained by NBL’s most frequent organs of origin – the adrenal glands / sympathetic paravertebral chain, and their adjacency to the major abdominal vessels.
The remainder of the IDRF present in the sample were insufficiently frequent to qualify for inclusion in the “most frequent” category.

This distribution in frequency can be used as a base for further analysis of IDRF characteristics on a much larger scale for the purpose of developing specific strategies for screening and treatment optimization.

In 2009 the International Neuroblastoma Risk Group reported that the effect of individual image defined risk factors is unknown. In relation to this, through the application of several statistical methods, we discovered several connections between patient survival and specific IDRFs:

By means of logistic regression analysis an association between the most frequent image defined risk factor in the sample (involvement of the renal pedicle) and survivability was revealed – the likelihood of patient survival is higher in the absence of said factor. This result could be rationalized by taking into consideration the critical homeostatic function of the kidney in the long term, as well as its ample vascularization in terms of intraoperative hemorrhagic complications in the short term.

Logistic regression analysis was applied when studying the additional condition pleural effusion. Similarly to the infiltration of the renal pedicle, the sample data showed an association between the outcome and present effusion – the patient is more likely to survive in the absence of said condition. These findings are concomitant with those of Cowie et al. 1997, Kammen et al. 2001, and Monclair et al. 2009 – they all link pleural effusion in neuroblastoma patients with decreased survivability. However, it must be pointed out that pleural effusion does not constitute an IDRF, nor metastatic disease by itself.

Next, dispersion analysis was applied to the number of IDRFs a patient has in relation to Event-Free Survival (EFS). When considering only the patients with a single image defined risk factor, the number of IDRFs demonstrated significance for changes in EFS with satisfactory accuracy. Simon et al. 2008 reports a similar finding, linking the absence of IDRF with better EFS values. In our study, the worst
survival rates were found in patients with the most IDRFs (maximum 3 in our sample).

Dispersion analysis was also applied to the data of all individual IDRFs in search of significance for Event-Free Survival. This revealed statistically significant prognostic value only for the factor ipsilateral tumor spread in two adjacent compartments.

In addition, Student’s t-test was used to examine for a difference between the distributions of living / deceased patients in cases with and without the individual IDRFs. From this a statistically significant difference in distribution was established in all IDRF, represented in the sample, save for one – infiltration of the renal pedicle. The image defined risk factors that manifest this difference in distribution are as follows:

- ipsilateral spread of tumor in two body compartments;
- tumor encasing brachial plexus roots;
- tumor encasing subclavian vessels and/or vertebral and/or carotid artery;
- tumor compressing the trachea (cervico-thoracic);
- tumor compressing the trachea and/or principal bronchi (thorax);
- lower mediastinal tumor, infiltrating the costo-vertebral junction between T9 and T12;
- tumor encasing the aorta and/or vena cava (thoraco-abdominal);
- tumor encasing the origin of the coeliac axis, and/or of the superior mesenteric artery;
- tumor encasing the aorta and/or vena cava;
- pelvic tumor crossing the sciatic notch;
- intraspinal tumor extension;
- infiltration of adjacent organs/structures (pericardium, diaphragm, kidney, liver, duodeno-pancreatic block and mesentery).
A statistically significant difference in the distribution of the variables “living” / “deceased” was found in all additional conditions, represented in the sample, namely: multifocal primary tumor, pleural effusion, and ascites.

Concomitant with this data are the findings of Avanzini et al. 2017. Said study reports that encasement of the branches of the mesenteric artery is predictive of a poor surgical outcome. In addition, the same study finds that abdominal midline perivascular IDRFs are prognostic not only for surgical outcomes, but also for Event-Free Survival and Overall Survival. This data supports our findings of a statistically significant difference in the “living / deceased” distribution of patients with and without a specific IDRF, as well as a potentially prognostic value of the image defined risk factors.

The final part of our study focused on comparisons between different groups of patients, taken from the overall sample. Kaplan-Meier graphs and Cox’s F-test were employed to compare the survivability of patients with a single and without any IDRF, as well as patients with more than one IDRF. The analyses were carried out based on Event-Free Survival and Overall Survival values. The following statistically significant differences in survivability were found:

- between patients without IDRF and patients with a single IDRF – in favor of patients without IDRF;
- between patients without IDRF and those with one or more IDRF – once more in favor of those without IDRF;
- between patients with a single IDRF and those with more than one IDRF – in favor of those with more than one IDRF (this illogical result is due to the double number of patients with a single factor within the sample, when compared to those with more than one factor).

To supplement this, comparisons of the survival graphs of more than two groups at once were performed using Log-Rank test and the values of EFS and OS. A statistically significant difference between the patients without IDRF, the patients with a single IDRF, and those with one or more was discovered in regard to OS. That
is to say that patients with a different number of IDRFs present at the time of diagnosis demonstrate different EFS and OS.

The results of Simon et al. 2008 are once more concomitant to our findings – the author reports a statistically significant correlation between the absence of IDRF and better Event-Free Survival values.

Monclair et al. 2015 concludes that in localized neuroblastoma IDRF at diagnosis are associated with worse survivability and a greater frequency of surgical complications – this is confirmed by our own results. Monclair suggests that the effect of IDRF be included in therapeutic planning.

The conclusions of the study conducted by Avanzini et al. in 2017 are similar to our results from the Kaplan-Meier comparative analyses – according to Avanzini full resection or minimal residual mass (ergo better pronosis) had been more frequently achievable in children with a reduced number of IDRFs / complete disappearance of IDRF post preoperative chemotherapy. Günther et al. 2011 makes similar rationalizations – the author finds a statistically significant negative correlation between the number of IDRFs and the possibility of full exeresis. The assumption can be made that the lower number of image defined risk factors correlates with a better prognosis.

Pohl et al. 2016 also reports of a negative association between the degree of macroscopic excision and the IDRF status, noting a link between the number of IDRF and the lower probability for radical resection. Penazzi et al. 2017 confirm this. These findings support the relations between the number of IDRF and the patient’s survival, discovered in our own study.

As part of the study a multivariate regressive Cox analysis was carried out on all available data to search for potential predictors prognostic of EFS and OS. This revealed that the INRGSS stage is significant for EFS survival.
VI. CONCLUSIONS

1. The most common IDRFs in the sample have been found – infiltration of the renal pedicle, intraspinal extension with compression, encasement of the abdominal aorta / vena cava and of the large mesenteric branches.

2. The most frequent IDRF (involvement of the renal pedicle) and the additional condition pleural effusion are associated with poor survival.

3. Individually all IDRFs and additional conditions present in the sample affect survivability.

4. There are differences between the survivals of patients without IDRFs, patients with one IDRF, and patients with more than one IDRF.

5. The presence of IDRFs worsens patient survival.

6. The patients with poorest survivability are those with the largest number of IDRF (up to a count of 3 IDRF in a single patient in the current sample).