Proposal Title: **Characterization of biologic features and outcomes in neuroblastoma patients with differentiating neuroblastoma histology**

Principal Investigators: **Elizabeth Sokol, MD, Ami V. Desai, MD, MSCE, Wendy London PhD, Sam Volchenboum, MD, PhD, and Susan Cohn, MD.**

Institution: **The University of Chicago**

Email address: esokol@peds.bsd.uchicago.edu, adesai12@peds.bsd.uchicago.edu, wendy.london@childrens.harvard.edu, svolchen@peds.bsd.uchicago.edu, scohn@peds.bsd.uchicago.edu

**Specific Aims:**

Neuroblastoma histology has been described as favorable or unfavorable based upon the International Neuroblastoma Pathology Classification (INPC) system. INPC includes grade of differentiation, mitosis-karyorrhexis index (MKI), and age. This criterion predicts outcomes well despite stage of disease. Age is an independent predictor of outcome and is used to help determine risk status outside of histology as well. Patients with differentiating disease compared with those with poorly differentiated or undifferentiated neuroblastoma are known to have better outcomes. We propose to better characterize the biologic features of this cohort of patients as well as the clinical outcomes. We will also evaluate the treatment that these patients have received in order to better target the best therapy for the patient without over- or under-treating.

**The specific aims of this proposal are:**

1) To use the INRG database (INRGdb) to better understand the tumor biology as well as clinical features associated with differentiating neuroblastoma.

2) To characterize outcomes associated with differentiating histology in order to identify optimal treatment.

**Hypothesis:**

We hypothesize that patients with neuroblastoma with differentiating histology have more favorable biologic features than patients with poorly differentiated or undifferentiated histology. In turn, we hypothesize that the clinical outcomes for these patients are better, indicating that these patients may not require high-risk therapy, despite their age. We will better refine risk prediction in patients who are older than 5 years who may not have high-risk disease.

**Patient Cohort (Eligibility Criteria):**

The analytic cohort will consist of the subset of patients in the INRGdb who have grade of histology information available. We will evaluate the age, treatment, stage, MYCN status, ploidy, chromosomal aberrations, disease sites, histology, diagnostic category, grade of differentiation, MKI, event free survival (EFS), and overall survival (OS). We will also evaluate the available demographic information.

**Background**

To ensure that therapy is appropriately tailored for patients with neuroblastoma, efforts to identify variables that accurately predict outcome have been ongoing for more than 35 years. Dr. Shamada defined histologic categories with grade of differentiation defined for patients with neuroblastoma. Differentiating histology was defined in patients with between minimal and 50% Schwannian stroma. A pathology-based risk stratification was defined by the International Neuroblastoma Pathology Classification (INPC). This system utilizes age at diagnosis, grade of differentiation, and MKI to define histologic risk group. An International Neuroblastoma Risk Group (INRG) Classification System has since been established based
on the statistical analysis of 36 prognostic factors in more than 8,800 patients diagnosed around the world. Using this system, patients are classified as very low-, low-, intermediate-, or high-risk according to the status of 7 prognostic variables. In lieu of using INPC, which stratifies tumors as unfavorable vs favorable histology according to patient age and tumor histologic features, three separate histologic criteria (diagnostic category, MKI, and grade) were evaluated to prevent the confounding of age as a prognostic criterion. Despite this, INPC histology continues to be utilized by the Children’s Oncology Group (COG) to determine treatment regimen.

Currently, patients who are over 5 years of age with neuroblastoma will receive high-risk therapy regardless of histologic grade because of their age. With INPC classification, any patient over 5 years has unfavorable histology. This means that a patient with differentiating histology may be high-risk based upon double counting of age. In the initial analysis of the 8800 patients within the INRGdb, there were 518 patients with differentiating histology. There were 2759 with undifferentiated histology. Both EFS and OS were significantly different between the two groups with $p < 0.0001$. EFS at 5 years was 83% in patients with differentiating histology and 63% in patients with undifferentiated histology. Similarly, 5-year OS was 89% in patients with differentiating histology, and 72% in patients with undifferentiated histology.

Evaluating the data a different way, patients with stage 3 disease were evaluated using the INRGdb. The 97 patients with differentiating histology had 5-year EFS and OS of 87% and 93% respectively. This is significantly longer than the 494 patients with undifferentiated histology who had 5 year EFS and OS of 74% and 81% respectively. Again, we see that patients with differentiating histology have better outcomes than patients with undifferentiated histology.

These comparisons do not separate this group of patients by age. With increased numbers of patients in the INRGdb, we can now more effectively characterize this subset of patients who are over 5 years and with differentiating histology to determine what is likely to be the optimal therapy for this group.

**Significance:**

Our overall goal is to ensure that each child with neuroblastoma receives treatment that is optimally stratified according to predicted risk. Unfortunately, currently there are some children who receive too much treatment, and others who receive too little treatment, due to errors in the risk stratification algorithm. By more careful evaluation of the patients with differentiating histology, we hope to identify a subset of patients who may be receiving more therapy than they need. We will better understand the biologic features of this subset of neuroblastoma tumors. We will also identify any demographic differences noted in this subset of patients.

We will additionally look at the treatment that these patients have received in combination with their event free and overall survival data. This will help us to determine whether this subset of patients do require high-risk therapy, or if they will have comparable clinical outcomes with intermediate-risk therapy. This could spare this subset of children the toxicity associated with autologous stem cell transplant, radiation, and immunotherapy. The INRGdb allows us to answer this question due to the large number of patients with data available.

**Experimental Approach/Methods**

**Proposal Description:**

We will pe...
• Site of primary tumor
• Among patients with metastatic disease, sites of metastases
• Primary tumor size (maximum diameter; to be analyzed as continuous variable and possibly dichotomized above and below the group mean)
• LDH at diagnosis (dichotomized at 587 U/L, per INRG standard)
• Ferritin at diagnosis (dichotomized at 92 ng/mL, per INRG standard)
• Urine catecholamines at diagnosis (positive vs. negative)
• MIBG avidity at diagnosis, if available
• Treatment received
• Treatment era

Biologic Features
• MYCN status (amplified vs. non-amplified)
• DNA index/ploidy (diploid vs. hyperdiploid)
• LOH at 1p (present vs. absent)
• Gain of 17q (present vs. absent)
• 11q aberration (present vs. absent)
• Pooled segmental chromosomal aberration (LOH at 1p, gain 17q, and/or 11q aberration vs. none of these changes)
• MKI (dichotomized as high vs. low-intermediate)

Clinical Outcomes
• Time to first event (or last follow-up if no event)
• Time to first relapse (or last follow-up if no relapse)
• Time to death (or last follow-up if no death)

Aim 1: To use the INRG data to better understand the tumor biology as well as clinical features associated with differentiating neuroblastoma.
We will identify all of the patients in the INRGdb with undifferentiated, poorly differentiated, or differentiating neuroblastoma. We will compare each of the biologic and demographic features between differentiating neuroblastoma and the other two groups in order to understand the characteristics of the differentiating tumor in patients older than 5 years.

To compare, we will use unpaired t-tests or rank sum tests for continuous variables. For categorical variables, we will use chi-squared tests. We will also perform multivariate analysis using Cox regression.

Aim 2: To characterize outcomes associated with differentiating histology in order to identify optimal treatment.
We will collect both EFS and OS data for patients with undifferentiated, poorly differentiated, and differentiating neuroblastoma. We will again compare the data for differentiating neuroblastoma patients to the other two cohorts. We will then analyze the treatment that these patients received. We will look at what percentage of these patients received high-risk and intermediate-risk therapy. We will compare the outcomes between the treatment groups. This will help to determine which type of therapy this unique subset of neuroblastoma patients requires.

We will use Kaplan-Meier analysis to estimate 5-year event-free, relapse-free, and overall survival rates according to grade of differentiation. We will use log-rank tests to compare outcomes between groups.
References


